THE FEEDING VALUE OF MANGELS.

BY T. B. WOOD, *Drapers Professor of Agriculture, Cambridge.*

THE series of experiments described in the following pages was commenced by Professor T. H. Middleton in the autumn of 1903. The experiments were designed to run side by side with an investigation on the chemical composition of mangels, the results of which appeared in this journal five years ago[1]. After Professor Middleton left Cambridge, the conduct of the experiments devolved upon the present writer.

Perhaps the most important point brought out in the paper referred to was the great difference in cropping power and chemical composition between the various types of mangels. The results bearing on this point are given in the following table.

TABLE I.

Name of type	Yield of roots per acre	Average content of dry matter	Yield of dry matter per acre
	Tons	%	Tons
White fleshed globe	29·9	10·7	3·2
Intermediate	27·4	12·0	3·3
Yellow fleshed tankard	24·6	13·1	3·2
Yellow fleshed globe	25·0	13·4	3·3
Long Red	29·9	13·1	3·9

The table, which embodies the results of a very large number of field experiments and analyses, shows clearly that the Long Red mangel produces about 20 per cent. more dry food per acre than any other variety. This being so, it would appear that the Long Red is far the most profitable mangel to grow.

It has, however, the disadvantage of being somewhat more troublesome to lift than the globes and tankards, since it buries its roots more deeply in the ground. But on this point the practical man is usually quite well informed. A much more important question is the

[1] Wood and Berry, Vol. I. Part 2, page 176.

reliability of the chemical determination of the percentage of dry matter as an index of feeding value.

Does it necessarily follow that the feeding value of any type of mangel is proportional to its percentage of dry matter?

It was to investigate this point that the experiments herein described were designed.

The plan of the experiments essentially consists in feeding side by side under similar conditions two lots of animals whose diets differ only in the variety of mangels supplied to them.

All the usual precautions as to selecting even lots of animals for experiment, weighing at equal intervals after food, and so on, were observed. About half the experiments were carried out on the University Farm at Impington. For the rest, Professor Middleton was fortunate enough to secure the co-operation of Mr E. R. Pratt of Ryston Hall, Downham Market, Norfolk, and Mr H. Shepherd Cross, M.P., of Hamels Park, Buntingford, Herts, to whom the sincere thanks of the author is due. But feeding experiments require constant supervision, and for this acknowledgments are due to Mr Henshaw, Manager of the University Farm, to Mr Newell of Ryston, and to Mr Rowley of Hamels Park. Without their constant attention, such experiments would have been impossible. Full details of all the earlier experiments, from Professor Middleton's pen, will be found in the *Guide* to Experiments of the Cambridge University Department of Agriculture for 1907. The points of interest for the present paper are given in the annexed table.

TABLE II.

No. of Expt.	1907 Guide Table No.	Year	Place	Kind of Animal	No. of Animals in each lot	Age of Animals, months	Duration of Experiment, days
1	XXII	1903–4	Impington	Fattening	4	36	84
2	XXIII	1904–5	,,	,,	6	15	168
3	XXIII	1904–5	,,	,,	3	25	84
4	XXVI	1906	Ryston	,,	6	32	117
5	XXVII	1907	,,	,,	6	30	112
6	—	1908	,,	,,	6	36	112
7	—	1909	,,	,,	6	36	42
8	XXIV	1905–6	Impington	,,	4	16	130
9	XXIV	1905–6	,,	,,	4	24	112
10	XXV	1907	,,	Stores	9	12	84
11	XXVIII	1904–5	Hamels Park	,,	8	26	117
12	XXIX	1905–6	,,	,,	8	33	92

The whole series of twelve experiments must be considered under three headings. In the first seven experiments the animals were fed on a liberal fattening ration, and the varieties of mangels compared were Long Reds and White-fleshed Globes, or Yellow Globes as they are commonly called. It will be convenient to deal first with the results of this comparison.

TABLE III.

Comparison of Long Red and Yellow Globe.

Expt. No.	Rations per head in lbs.				Average daily gain per 1000 lbs. live-wt. in lbs.		Increase due to Long Red, Yellow Globe = 100
	Cake and Meal	Hay	Straw	Roots	Long Red	Yellow Globe	
1	4–5	10	—	80—100	1·9	1·5	127
2	3—4	8	3	40	3·5	3·5	100
3	4—5	7	6	50	2·7	2·7	100
4	3	—	8	80–130	1·7	1·5	112
5	3	—	8	80–130	2·6	2·0	130
6	3	—	8	80—120	2·1	1·5	141
7	3	—	8	80—130	1·7	1·4	121

In many cases the ration was increased during the experiment. This is indicated in the table. For instance, in the roots column, 80—100 means that the animals received at the beginning of the experiment 80 lbs. of roots per head per day, and that this was subsequently increased to 100 lbs. per head per day. The average daily gain is calculated on the initial live-weight. The figure was obtained by taking the average daily gain per 1000 lbs. initial weight of each individual.

The increases are given in 1000 lbs. live-weight in order to eliminate the variation due to the size of the animals. In the last column the increases due to Long Red are recalculated, taking in each case the increase produced by Yellow Globe as 100. This makes it possible to compare the figures of different years and different ages of cattle more conveniently.

Inspection of the figures in this column shows that in no case did Long Red produce a worse result than Yellow Globe; in two cases out of seven the results were equal, and in five cases out of seven Long Red produced a distinctly better result than Yellow Globe. It seems there-

fore fair to conclude that the Long Red mangel has a higher feeding value than the Yellow Globe.

Feeding experiments, however, are liable to so many sources of error that some little consideration is necessary before definitely accepting this conclusion.

On examining the increases made by individual animals, for instance the four animals fed on Long Red mangels in Experiment 1 as given in Table XXII[1], the following results are obtained.

TABLE IV.

Animal No.	Monthly increase previous 10 months	Live-weight at beginning of experiment	Total gain in 84 days	Average daily gain per 1000 lbs. live-wt.	Average daily gain worst animal = 100
	lbs.	lbs.	lbs.	lbs.	
92	36·9	1276	249	2·3	152
94	38·3	1228	204	2·0	124
95	38·0	1190	182	1·8	112
91	40·2	1262	164	1·5	100

These figures show clearly that in spite of every precaution to choose for the experiment animals of uniform weight, and of uniform capacity for growth, the variation among the individual animals receiving the same treatment is far greater than the quantity the experiment seeks to measure. Thus the difference between the increase due to Long Red and that due to Yellow Globe is 27 per cent., as compared with individual differences of 52 per cent.

And this experiment is by no means an extreme case, even among the experiments described in this paper.

The question naturally arises: What kind of reliance can be placed on the average increase of a small number of animals whose individual increases differ to this extent? In the case of chemical or physical experiments, this would ordinarily be determined by calculating from the individual results the "probable error" of the average, and there seems to be no reason why this method should not be applied here. Proceeding in this manner, the following figures are obtained:

[1] Guide to Experiments, 1907.

TABLE V.

Experiment No.	Increase due to Long Red, Yellow Globe = 100	Probable Error[1]
1	127	±11
2	100	±9
3	100	±10
4	112	±8
5	130	±10
6	144	±16
7	121	±13

Experiments 2 and 3 are obviously of no significance. In Experiments 4 and 7 the differences in favour of Long Red are not much greater than the "probable error." In Experiments 1, 5, and 6, however, the differences are nearly three times as great as the probable error, and they must be taken as showing a distinct superiority in feeding value in the Long Red mangel.

In combining such results it is usual to regard the probable error as a measure of the reliability of the average result, and to weigh each result appropriately, *i.e.* inversely as the square of the probable error, before taking the final average. The collective result of the seven

[1] The following formulae were used in calculating the figures of this and succeeding tables. If d = the deviation of an observed quantity (*e.g.* increase per 1000 lbs. live-weight), from the mean of n such quantities, then the probable error of the mean of the n quantities $= 0{\cdot}67 \sqrt{\frac{\Sigma d^2}{n(n-1)}}$.

Thus the probable error of the mean daily gain per 1000 lbs. live-wt. (1·9 lbs.) of the four animals included in Table IV. $= 0{\cdot}67 \sqrt{\frac{0{\cdot}4^2 + 0{\cdot}1^2 + 0{\cdot}1^2 + 0{\cdot}4^2}{4 \times 3}} = 0{\cdot}1$.

The mean daily gain per 1000 lbs. live-weight would then be written $1{\cdot}9 \pm 0{\cdot}1$ lbs. which means that the true daily gain per 1000 lbs. live-weight of animals fed under the conditions of this experiment would be equally likely to fall outside the limits of $1{\cdot}9 + 0{\cdot}1$ lbs. and $1{\cdot}9 - 0{\cdot}1$ lbs. as inside those limits. For calculating the probable error of the ratio of the increase due to Yellow Globe to that due to Long Red, the formula used was

probable error of ratio $\frac{a}{b} = \frac{1}{b} \sqrt{p_a^2 + \frac{a^2}{b^2} p_b^2}$,

where p_a and p_b are the probable errors of a and b.

I am indebted to Mr F. J. M. Stratton and Mr A. B. Bruce for the above and other similar paragraphs.

experiments becomes then Yellow Globe : Long Red = 100 : 116 ± 4, the probable error being estimated from the deviation of the experiments amongst themselves[1].

According to the ordinary tables of probable errors, this means that the chances are 300 to 1 in favour of Long Red being better in feeding value than Yellow Globe, 30 to 1 that the difference in favour of Long Red is at least 5 per cent., and 5 to 1 that it is at least 10 per cent.

The final result shows that, taking the increase produced by a certain quantity of Yellow Globe mangels forming part of a liberal fattening ratio as 100, the increase which may be expected to be produced by an equal quantity of Long Reds under similar conditions is about 116.

It seems therefore justifiable to conclude from this investigation that the feeding value of the Long Red mangel is about 16 per cent. greater than that of the Yellow Globe mangel.

The average percentages of dry matter in the mangels used in the experiments were as follows:

TABLE VI.

	Dry matter per cent.		Dry matter in Long Red, Yellow Globe = 100
	Long Red	Yellow Globe	
Impington ...	13·1	10·7	122
Ryston	10·6	9·0	118

There is evidently a close agreement between the relative percentages of dry matter in these two types of mangels and their feeding value.

[1] Thus, if r is the probable error of each experiment, and $\frac{1}{r^2}$ consequently the weight given to that experiment in calculating the weighted mean, and if d is the deviation from the weighted mean, then the probable error of the weighted mean is

$$0{\cdot}67 \sqrt{\frac{\Sigma \frac{d^2}{r^2}}{(n-1)\,\Sigma \frac{1}{r^2}}}.$$

Calculated in this manner the ratio of the increase produced by Yellow Globe mangel as a constituent of a liberal fattening ration to that produced by an equal quantity of Long Red = 100 : 116 + 4.

Comparison of *Long Red* and *Yellow-fleshed Tankard*, or *Golden Tankard* as it is usually called, for fattening cattle.

TABLE VII.

Expt. No.	Rations per head in lbs.				Average daily gain per 1000 lbs. live-wt. in lbs.	
	Cake and Meal	Hay	Straw	Roots	Long Red	Golden Tankard
8	2—4	8	—	60—84	2·3	2·5
9	3—5	8	4	90—112	1·4	1·5

The figures show that in these two experiments the two types of mangels compared produced practically equal results, the difference if any being well inside the probable error of such experiments. This being so, equality in feeding value is indicated, and this agrees well with the percentages of dry matter—Long Red 13·1 per cent., Golden Tankard 13·4 per cent. It did not seem worth while to pursue this question further.

Comparisons of *Long Red* and *Yellow Globe* for *Store* cattle.

TABLE VIII.

Expt. No.	Rations per head in lbs.				Average daily gain per 1000 lbs. live-wt. lbs.		Increase due to Long Red, Yellow Globe = 100
	Cake and Meal	Hay	Straw	Roots	Long Red	Yellow Globe	
10	2/3—2	3	7	50—100	1·3	1·2	108
11	1—3	3	7	50—90	1·7	2·1	81
12	1	—	3	40—56	0·9	0·9	100

In Experiment 11 the plan was slightly varied. An attempt was made to adjust the diets of the two lots to practical equality by adding to the diet of the Yellow Globe lot enough maize meal to make up for the deficiency in dry matter of the poorer mangels. The Yellow Globe mangels contain an appreciably higher percentage of nitrogenous substance than Long Reds. In spite of this substitution of maize meal for part of the cake in the case of the Yellow Globe lot, this lot actually received more nitrogen in their daily ration, and not only that, but their ration had a higher albuminoid ratio, 9·6 as compared with 10·2. Both

these are wide ratios, and the extra nitrogen provided by the Yellow Globes may account for the greater growth of these animals.

In the other two experiments the differences observed are inside the probable error of such experiments. The result of these three experiments with animals on a store diet is therefore inconclusive, but suggests that possibly Long Red mangels may not have the same value in a store diet as they have in a liberal fattening ration rich in nitrogen.

Summary.

The paper describes attempts to test the following points—the comparative feeding value of Yellow Globe and Long Red mangels as constituents of a liberal fattening diet, the comparative feeding value of Golden Tankard and Long Red mangels, also as constituents of a fattening diet, and the comparative feeding value of Yellow Globe and Long Red mangels for store cattle.

The results point to the following conclusions:

The rates of fattening of individual animals vary so greatly that little reliance can be placed on the results of single experiments with the small numbers of animals commonly employed in feeding tests.

The feeding values of Long Reds and Yellow Globes were compared on seven occasions, and the results discussed according to the ordinary methods used in the theory of probabilities.

From the final result it is concluded that the relative feeding values of these two types are approximately as 116 : 100 in favour of Long Red.

This result agrees as well as can be expected with the relative percentages of dry matter, which are as 120 : 100, also in favour of Long Red.

Two comparisons of Long Red and Golden Tankard indicated that there is no appreciable difference in the feeding value of these types. This also agrees with the fact that their percentages of dry matter are practically equal.

The three experiments with store cattle are regarded as inconclusive.

THE AMMONIA IN SOILS.

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It has long been recognised that the amount of ammonia in soils cannot be determined by distillation with a solution of caustic soda or potash since these strong alkalis slowly decompose the complex nitrogenous matter and evolve a continuous stream of ammonia. Boussingault showed more than fifty years ago how this difficulty could be obviated. By using magnesia in place of soda or potash the decomposition of such substances as urea, asparagin, and albumin was not great even on long boiling, but when the distillation was carried out under reduced pressure at 38°—40° there was no decomposition at all[1]. Ammonium salts, on the other hand, were completely broken up in these circumstances. He used this low pressure method in his researches on the ammonia content of urine[2], but does not appear to have applied it to soils. Sufficiently accurate results could, he considered, be obtained by distillation at 100°, and this method was for many years generally adopted. It gave results varying according to the nature of the soil from 10 to 100 parts of nitrogen per million (·001 to ·01 per cent.), which may still be found quoted in some of the agricultural text books.

Evidence was, however, obtained later that Boussingault had underestimated the decomposing effect of boiling aqueous magnesia on nitrogen compounds, and two other methods are attributed by Grandeau to Schloesing[3]. One suitable for approximate determinations consisted in leaving a mixture of the soil with strong caustic soda solution under a bell jar in which was also placed some standard acid to absorb the ammonia evolved. The other was recommended where greater precision was necessary; it not only lessened the risk of decomposition but also aimed at getting over another difficulty that had been

[1] "Du dosage de l'ammoniaque en présence des substances organiques azotées," *Agronomie*, 1864, III. 206.

[2] "Recherches sur la quantité d'ammoniaque contenue dans l'urine," *ibid.* p. 233. Lime was used in these experiments in place of magnesia.

[3] Grandeau, *Analyse des Matières Agricoles*, 1879.

recognised—the re-absorption of the liberated ammonia by the soil. 100 grams of soil are treated with dilute hydrochloric acid till all the carbonates are neutralised and the liquid remains distinctly acid; water is added and the mass is shaken. The liquid is now supposed to contain all the ammonia; an aliquot portion is therefore filtered and distilled in the ordinary way with potash. The hydrochloric acid must obviously be free from traces of ammonia and has usually to be specially re-distilled from a little sulphuric acid.

In their investigations on the nitrogen compounds of the soil Berthelot and André made use of the following method[1]:—100 grams of soil are heated for 30 hours on a steam bath with about 500 c.c. of water and 15 grams of hydrochloric acid. The liquid is then filtered and used for the determination of ammonia.

A method quite distinct in character was devised by W. Wolf and S. Knop[2]. The soil was treated with sodium hypobromite which reacts with ammonia producing nitrogen. Baumann[3] has, however, shown that the volume of gas evolved is not a true measure of the amount of ammonia present; more consistent results are obtained if a hydrochloric acid extract of the soil is first oxidised with ozone and then treated with the hypobromite.

None of these methods is free from convention; it is always assumed that the ammonia or nitrogen evolved during the operation represents what is actually present in the soil as ammonia, but no proof of this assumption is ever offered. In the present state of our knowledge, however, no method can be entirely free from convention; nothing short of the actual isolation in the pure state of the various nitrogen compounds in the soil would enable one to say definitely what is and what is not an ammonium compound. It is necessary to agree on some definition; for the purpose of this paper a substance is called an ammonium compound if it evolves ammonia rapidly, completely, and in one stage when treated with weak alkalis at low temperatures.

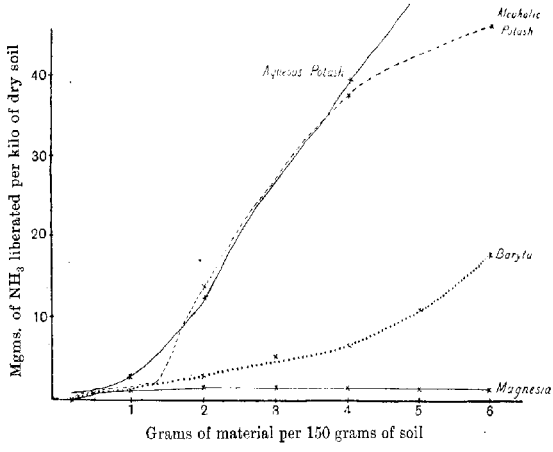
The action of alkalis on the soil.

The amounts of ammonia evolved from soil on distillation at 38°—40° with various alkalis is given in Table I, and the results are plotted on the curves in Fig. 1. It is clear that the action is not the same in all cases. The quantity of ammonia given off by baryta increases

[1] *Annales de Chim. et de Phys.* [VI] XXV. 327.
[2] *Chem. Centralblatt*, 1860, pp. 243 and 534.
[3] *Landw. Versuchs-Stationen*, 1886, **33**, 247.

continuously as the quantity of baryta increases and there is no sign of a break in the curve. In every instance, even when only 0·5 gram

Soil 1. Poor arable soil



Soil 2. Dunged soil

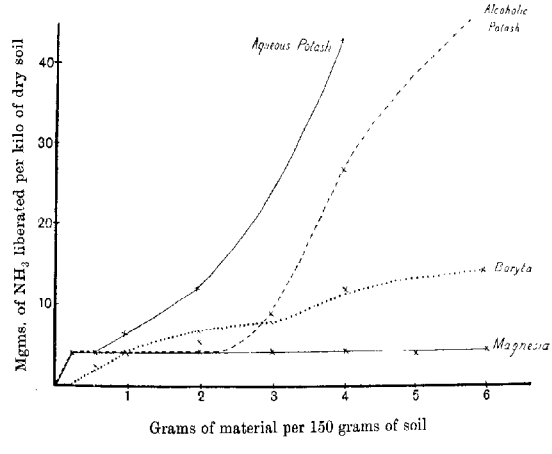


FIG. 1.

is present, there is much more baryta than is required to liberate the ammonia if the reaction were simply between the baryta and an ammonium salt. The result indicates that baryta is causing some decomposition of other nitrogenous compounds with formation of ammonia.

Aqueous potash behaves like baryta, but the decomposing action is more vigorous.

The amount of ammonia liberated by varying quantities of magnesia does not rise continuously; it increases for a short time and then remains constant.

There is, however, some decomposition. If at the end of the distillation a fresh quantity of water is added and the distillation continued for a second period there is a further evolution of ammonia thus:

Quantities of magnesia in grams per 150 grams of soil	0·2	0·5	1·0	2·0	3·0	4·0	5·0	6·0
Ammonia evolved, mgms. per kilo of soil,								
1st distillation.....	3	3	4	4·5	4	4·5	2·5	4
2nd „	2·5	5	5	4·5	5·5	5·5	5	4·5
3rd „	2	—	—	6	5·5	7	—	—

The difference in action between magnesia and baryta is thus rather one of degree than of kind. The difference in the curve arises from the fact that successive increments of baryta produce solutions of increasing concentration, while magnesia, on account of its low solubility, forms in all cases a solution that is saturated, and therefore constant in concentration.

Alcoholic potash, on the other hand, gives a curve of altogether different character. The action is divided into two stages; in the first, where less than two or three grams of potash is present per 150 grams of soil, a constant quantity of ammonia is given off and the curve of decomposition is a straight line; in the second, where more potash is present, the evolution of ammonia increases with each increase in the amount of potash.

The first stage was found to represent the decomposition of a definite compound or group of compounds distinctly marked off from the other nitrogen compounds in the soil. The proof lay in the fact that no further evolution of ammonia took place on adding fresh quantities of alcohol after the first distillation was over and continuing the distillation for a second period. It was found that there was always an excess of potash (except perhaps when only 0·2 gram was used) to effect decomposition had any decomposable compounds still remained. But in the second stage some other compounds are attacked, and the

decomposition instead of coming to a sharp end continues indefinitely. The results are as follows:

Quantities of potash (in alcohol) in grams per 150 grams of soil		0·2	0·5	1·0	2·0	3·0	4·0	5·0	6·0
Ammonia evolved, mgms. per kilo of soil,			1st stage						
	1st distillation.....	3	8	9·5	9·5	23·5	34	—	—
	2nd ,, ...	0	0	0	2·5	7	9	—	—
			1st stage						
Another soil,	1st ,, ...	3	3	3	5·5	8·5	22·5	35	41·5
	2nd ,, ...	0	1	0	1	4·5	10	19·5	19·5

Turning now to the results obtained with the various soils it will be noticed that the arable soils give off as much ammonia under the action of small quantities of alcoholic potash as of magnesia.

No. of soil	4	5	6	7	
Ammonia evolved by magnesia (2 grams)	1	2	2·5	1·5	
Ammonia evolved by alcoholic potash (0·7 gram)	2	2·5	2	1	mgms. per kilo

This is generally true of arable soils and indicates that the amount of continuous decomposition effected by magnesia is inappreciable under the conditions of the experiment. Otherwise such concordant results are not obtained; if, for instance, the magnesia distillation is continued for some hours after the mixture has gone dry, and particularly if the temperature of the bath rises during the period, there is a continuous, though small, evolution of ammonia. An example is given in Table I, Soil 2.

TABLE I. *Ammonia evolved from various soils when distilled at low pressure with solutions of alkalis.*

Soil 1. Arable soil containing ·178 per cent. of nitrogen and losing 4·57 per cent. on ignition.

Alkali used	Mgms. of ammonia evolved per kilo of soil when 150 grams of soil are distilled with 100 c.c. of water (or alcohol) and the following amounts of alkali in grams								Temperature of distillation
	·2	·5	1·0	2·0	3·0	4·0	5·0	6·0	
Aqueous potash	1	2	3	12·5	—	40	—	65	38°
Baryta	0	0·5	1	3	6	7	11·5	18	38°
Magnesia	0	0·5	1	1·5	2	1·5	1·5	1·5	38°
Alcoholic potash	0	1·5	1·5	14	—	38	—	47	26°

Soil 2. Well dunged arable soil (14 tons of dung annually) containing 0·256 per cent. of nitrogen and losing 8·88 per cent. on ignition.

Alkali used	Ammonia evolved when the following grams of alkali are used								Temperature of distillation
	·2	·5	1·0	2·0	3·0	4·0	5·0	6·0	
Aqueous potash	3	4	6	12	—	43	—	77	38°
Baryta	0	1·5	4	7	8	13	11·5	16	38°
Magnesia	4	4	4	6	4	4	3·5	4	38°
Alcoholic potash	4	2	4	4	9	16·5	50	—	26°
Magnesia	6	5·5	7	7	7	10	8·5	7	45°
Magnesia	4	9	12	11·5	10	11	11·5	10	kept all night at 45°

Soil 3. Pasture soil containing 0·318 per cent. of nitrogen and losing 9·94 per cent. on ignition.

Aqueous potash	3·5	11·5	13·5	15·5	—	23	—	60	38°
Baryta	3	4·5	9	10·5	—	16·5	—	37·5	38°
Magnesia	4·5	5	5	5·5	—	5·5	—	6	38°
Alcoholic potash	1·5	2	2·5	4·5	9	14·5	34	40	26°

It is difficult to ascertain definitely whether all the ammonia actually evolved has been collected in the receiver or whether some has been physically re-absorbed by the soil. Under normal conditions soil is known to have considerable power of absorbing ammonia, but there is nothing to show that it can do so under the conditions of the experiment, *i.e.* when 100 grams of liquid are volatilised from 150 grams of soil at a temperature of 38° under a reduced pressure of some 10 mm. only. Indeed the indirect evidence is all the other way. When the amount of water distilled from the soil was increased from 100 to 250 c.c. there was a slightly increased evolution of ammonia such as might have been expected from the decomposition known to go on, but not sufficient to indicate that any notable absorption had taken place when only 100 c.c. were used. A typical result was as follows:

c.c. of water added to soil (150 gms. + 2 gms. MgO) and distilled off completely	100	150	200	250
Ammonia evolved, mgms. per kilo of soil	9	9·5	10	10·5

Further, wood-charcoal was found to possess a greater ammonia-absorbing power than soil, yet it only retained about 15 per cent. of the added ammonia on distillation with ammonium salts in quantities comparable with those present in the soil.

The results so far obtained may be summarised as follows:

(1) When soil is distilled at low pressure with dilute alcoholic potash (·5 to 1 or 2 of potash in 100 of alcohol) a definite group of nitrogen compounds is decomposed with evolution of ammonia. The action comes to an end as soon as these compounds are broken up.

(2) When aqueous potash, baryta, magnesia, or more concentrated solutions of alcoholic potash are used the action does not come to a sharp end but continues indefinitely. Magnesia, however, only shows to a small extent this subsequent decomposition and evolves during the course of one distillation the same amount of ammonia as the small quantities of alcoholic potash.

(3) The nitrogen compounds thus marked off from the rest by small quantities of alcoholic potash and, less sharply, by magnesia are regarded as ammonium salts because they are a distinct group yielding up their ammonia rapidly and completely. They include the ammonium compounds in the soil (for pure ammonium salts readily decompose under these conditions) and if any others are present they are so like ammonium compounds that no inconvenience can arise through classing them all together.

(4) There is no reason to suppose that soil physically absorbs much ammonia during the process.

The determination of ammonia in soils.

The two following methods have been worked out on the basis of these results.

(1) *Distillation with magnesia.* 150 grams of the soil are distilled at low pressure with 2 grams of magnesia suspended in 100 c.c. of water; the ammonia evolved is collected in standard acid, N/100 by preference, and estimated by titration. In order to ensure the purity of the reagents the magnesia is first introduced into the distillation flask (one of 500 c.c. with a tap passing through the stopper answers satisfactorily), 100 c.c. of water are added and a label is affixed to mark the level, then another 100 c.c. are added and the liquid boiled till the original level is again reached. Then cool, add the 150 grams of soil, attach the flask to the receiver, which consists of an ordinary stout filtering flask. No condenser is necessary, but it is an advantage to use a 100 c.c. pipette as the connecting tube in case any of the acid should run back. The distillation flask is now put into a water bath at 40°, attached to the water pump, and left for six hours, when all the

liquid should have passed over. Air can now be admitted through the tap in the stopper and the flask disconnected; the pipette is washed out and the acid titrated. The apparatus is shown in Fig. 2.

(2) *Distillation with alcoholic potash.* 0·7 gram of *freshly fused* potash and 150 c.c. of alcohol are introduced into the flask and 50 c.c. of alcohol distilled off; after cooling the soil is added and the distillation conducted as before excepting that the bath is to be kept at 25° only. The alcohol must be removed from the distillate before titration

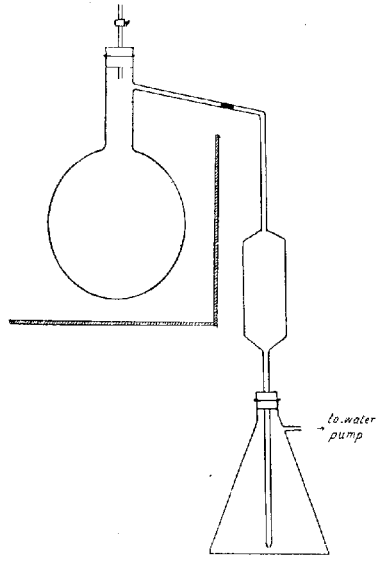


FIG. 2. Showing half of water bath with one set of apparatus for determining ammonia in soils, there being another set in the other half.

or it interferes with the sharpness of the reaction; the liquid is therefore first heated for an hour on the water bath.

The first of these methods is identical in principle with one devised by Boussingault and revived many years afterwards by Longi; more recently it has been adopted by Horace Brown in investigating the nitrogen compounds in malt extract. It is by far the more convenient of the two, and has been in regular use at Rothamsted for over twelve months with very satisfactory results. Its drawback is that it

necessitates the soil being kept at 40° for some time, at which temperature some decomposition has been found to take place. The second method is more troublesome to work, but only requires a temperature of 25° and does not appear to be complicated by any decomposition.

In all cases as yet tested the two methods give identical results provided the temperature does not rise too high. It appears, then, that the amount of decomposition effected by magnesia during the six hours of the distillation is too small to influence the result. The magnesia method has therefore been adopted in the experiments described below because of its greater convenience in working.

Duplicate determinations usually agree to within one part of nitrogen per million of dry soil. Most of these experiments have, however, been made with arable soils, only a few with garden and pasture soils, and none at all with peaty soils.

The effect of adding ammonium compounds to soil.

The method described above was, as a test, applied to soils containing an admixture of ammonium salts in known amounts. Many trials were made, but in no case was it possible to recover the whole of the added ammonia; a certain amount invariably entered into some combination from which it could not be expelled by any alkalis under the conditions of the experiment. A measured quantity of standard ammonium chloride was added to the mixture of 150 grams of soil, 2 grams of magnesia and 100 grams of water in the distillation flask; the whole was shaken for a few minutes and then distilled. Only

Soil used	Alkali	Ammonia added per 150 grams of soil (expressed as nitrogen)	Ammonia recovered (expressed as nitrogen)	Percentage entering into stable combination
		grams	grams	
Stiff arable loam	Magnesia	·001	·0005	50
	,,	·0009	·0006	33
	Baryta	·001	·0007	30
Well dunged stiff arable	Alcoholic potash	·0025	·0013	50
Pasture soil	,,	·0025	·0012	50
Rich garden soil	Magnesia	·0005	·0003	40
Subsoil (stiff clay)	,,	·001	·0006	40
	,,	·003	·0022	28
	Baryta	·001	·0006	40
	,,	·003	·0023	24
Modelling clay	Magnesia	·0025	·0019	25
Sand	,,	·0025	·0022	14
150 sand + 5 natrolite	,,	·0025	·0022	14

about 50 to 70 per cent. of the added ammonia was liberated. It seems unlikely that bacteria could, in the short time of the experiment, have taken up the rest, and physical absorption seems equally improbable in view of the enormous ratio of water volatilised to ammonia added (10,000 : 1 or 3). It is well known that an insoluble substance is formed when ammonium salts are added to soil, which only slowly becomes available to plants. The stability of this compound towards alkalis under diminished pressure indicates that it is not a true ammonium salt, but belongs to some other type of nitrogen compound. Its nature has yet to be determined.

Experiments on the rate at which it is nitrified in the soil have so far led to inconclusive results. That it does not indefinitely accumulate in the soil is indicated by the statistics of the Rothamsted plots annually receiving ammonium salts as manure; the total nitrogen does not tend to increase as years go on, but rather to decrease.

The amounts of ammonia found in certain soils.

In Table II are given the amounts of ammonia found in various Rothamsted soils and subsoils at different times of the year. The unmanured plot contained in April 1·6 parts of ammonia per million of soil, or about 3½ lbs. per acre in the top 9 inches and rather smaller quantities during the summer; by October it had practically disappeared. The dunged plot—which had received its manure seven weeks previously—contained seven parts of ammonia but a month later only four parts were found, or 9 lbs. per acre in the top 9 inches, and there was no change in this amount throughout the season. The Hoos Field plots 1A, 2A, and 4A had received their dressings of ammonium salts on Feb. 20 and 21st, and there had been 3·9 inches of rain during the seven weeks before April 8th, the date of sampling, yet a considerable amount of ammonia could be detected, at any rate on plot 4A. This is in accordance with the well known fact that ammonium salts do not wash out of a soil by rain. But during the next four weeks when the weather was warmer the ammonia dropped to two parts per million only, although only 1·85 inches of rain had fallen; it was not washed out in wet weather but it was rapidly nitrified when the soil became dryer and warmer. On the Broadbalk field the bulk of the ammonium salts had been applied only the day

TABLE II. *Nitrogen as ammonia in one million parts of soil dried at 100° C.*

Surface soils.

Date of sampling	Hoos Field Barley plots					Broadbalk wheat plots		Arable soils in fair condition cultivated fallow		
	Dung[1] Plot 7–0	Unmanured Plot 1–0	Ammonium salts only[2] Plot 1 A	Ammonium salts and superphosphate Plot 2 A	Ammonium salts and complete minerals Plot 4 A	Ammonium salts only Plot 10	Ammonium salts and complete minerals Plot 7	1	2	3
1909										
April 8	7·0	1·6	4·3	—	13·0	12·9	18·6	1·3	1·0	0·7
May 7	4·0	—	1·6	1·6	2·0	2·6	15·0	2·0	2·0	1·6
June 11	4·0	1·0	1·6	1·6	1·6	2·2	2·2	1·6	2·0	1·0
July 12	5·3	1·0	1·6	1·0	2·2	1·6	4·3	1·0	1·6	1·0
Oct. 28	4·0	·5	1·0	1·0	1·0	1·6	2·2	1·0	trace	2·2
Subsoils.										
Oct. 28	nil	nil	nil	nil	nil	nil	trace	trace	nil	nil

[1] The dunged plot receives 14 tons of dung each year and has done so continuously since 1852.

[2] Plots 1 A, 2 A and 4 A receive 200 lbs. of ammonium chloride and sulphate (mixed) containing 43 lbs. of nitrogen.

before[1]. Ammonia quickly disappears from plot 10 and is gone by the end of the month, but it persisted for some long time on plot 7. The other soils never contained more than about two parts of ammonia per million and this number was fairly constant throughout the whole season falling somewhat, however, in October. No measurable amount of ammonia was ever found in the subsoil. Many other soils have been examined at various times with the same results.

The important fact brought out by these results is that the ammonia in the soil tends to remain at a constant minimum depending on the quantity of organic matter present. Ordinary arable soils contain one or two parts per million while rich dunged soils and garden soils contain three or four. Subsoils contain none.

It follows that under ordinary conditions nitrification goes on more rapidly than ammonia production, even on the rich dunged soil; if it did not ammonia would accumulate. The rate of nitrification is, in consequence, limited by the rate of ammonia production.

[1] Three-quarters of the dressing was applied on April 7th, the other quarter having been put on on the previous Oct. 7 and 8.

This observation accounts for a number of discrepancies that have come to light in the various experiments on the rate of nitrification in soils. Determinations have been made in some instances of the rate at which nitrates accumulate in the soil, in others of the rate at which nitrates are produced in culture solutions containing ammonium salts inoculated with some of the soil. It has been customary to suppose that both methods deal with the same thing and should therefore give comparable results; we now see, however, that they deal with totally different things and it is not surprising that the results do not agree[1]. The rate of accumulation of nitrates in the soil measures the *rate of ammonia production in the soil*; the production of nitrates in a culture solution, or the "nitrifying power," measures the *rate of nitrification in a solution*. In order to get the *rate of nitrification in soil* it would be necessary to work with mixtures of soil and ammonium salts.

Although no accumulation of ammonia in the soil takes place under normal natural conditions it can be brought about by artificial means. When the nitrifying organisms are killed by heat or by toluene, as in the experiments recently described by the writer and Dr Hutchinson[2], the ammonia rises to 30 or 40 parts per million of soil. In the same paper is recorded another phenomenon of interest in this connection. The addition of toluene to soil causes an instant liberation of ammonia, amounting to 3 or 4 parts per million of soil; this behaviour is also shown by other volatile organic substances. W. E. Armstrong and E. F. Armstrong have recently shown that hydrocyanic acid is at once liberated when a leaf of cherry laurel is exposed to the vapour of an anaesthetic, and they have applied the name *hormone* to a substance which can thus get into a cell and hasten the normal katabolic changes. The two sets of phenomena are so similar that they can scarcely be disconnected. Further experiments on the sudden liberation of ammonia from soil are now in progress.

Conclusions.

(1) When soils are distilled at low pressures with small quantities of potash dissolved in alcohol a definite amount of ammonia is evolved and the reaction then comes to an end. It is considered that this amount represents the ammonium salts in the soil.

[1] Stevens, Withers, Temple and Syme have recently examined a number of soils by both methods and find no sort of agreement in the results (*Centr. für Bakt.* Abt. II. 1909, **23**, 355).

[2] This vol. p. 111.

When larger quantities of potash are used, or when baryta or magnesia in aqueous suspension is substituted, the decomposition is not definite but continues indefinitely. During the progress of the first distillation, however, magnesia gives off the same quantities of ammonia as small quantities of alcoholic potash.

(2) Two methods based on these observations are given for estimating the amount of ammonia in soils. If the amount of organic matter is not too high distillation with magnesia at reduced pressure gives accurate results, otherwise it is necessary to use alcoholic potash.

(3) The quantity of ammonia in samples of soil taken at different periods of the year is found to be constant but very small, being only about one or two parts per million of soil. The higher the amount of organic matter the greater the ammonia content, rising to five or six parts per million on heavily dunged arable or garden soils.

(4) As there is no tendency for ammonia to accumulate it follows that the rate of nitrification must be greater than that of ammonia production and in normal conditions is limited by this rate. Reviewing in the light of this observation the various methods of studying the rate of nitrification in soil it is seen that they really deal with three separate things—the rate of ammonia production in soil, the rate of nitrification in soil, and the rate of nitrification in a culture solution. In these circumstances it is not surprising that concordant results have not been obtained.

(5) When ammonium salts react with soil a certain proportion enters into a stable combination which is not decomposed on distillation with alcoholic potash or magnesia and is therefore not an ammonium compound. Its constitution has, however, not been determined.

THE EFFECT OF EARTHWORMS ON SOIL PRODUCTIVENESS.

By EDWARD JOHN RUSSELL.

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GILBERT WHITE devotes one of his letters to earthworms. "Worms," he says "seem to be the great promoters of vegetation, which would "proceed but lamely without them, by boring, perforating, and loosen- "ing the soil, and rendering it pervious to rains and the fibres of plants, "by drawing straws and stalks of leaves and twigs into it; and, most of "all, by throwing up such infinite numbers of lumps of earth called "worm-casts, which, being their excrement, is a fine manure for grain "and grass... the earth without worms would soon become cold, hard- "bound, and void of fermentation, and consequently sterile." Sixty years later, in 1837, Darwin published a paper[1], in which he showed the important part played by worms in the formation of vegetable mould. Further observations were recorded by Hensen[2] in 1877. Agricultural chemists did not, however, generally make use of any of this work, and it was not till 1881 that the publication of Darwin's *Earthworms and Vegetable Mould* directed so much attention to the subject that the action of earthworms could no longer be disregarded. Several investigations have since been made into the part played by earthworms in promoting fertility, perhaps the best known being those recorded in Wollny's *Zersetzung der organischen Stoffe*. Some of these dealt with the physical effects produced, particularly the loosening of the soil, others had reference to the production of plant food. Plants grown in boxes of soil containing earthworms made much better growth than others in soil from which earthworms had been removed, the increased yield amounting to 100 per cent. or more—in one case over 700 per cent.;

[1] *Trans. Geological Society*, **5**, 505.

[2] *Zeit. f. Wissenschaft. Zoologie*, 1877, **28**, 361.

chemical analysis of the soil showed a larger quantity of nitrogenous food stuff where worms were present:

	Nitrogen as		Sum
	Ammonia	Nitrate	
Soil + earthworms	·01647	·02204	·03851
Soil without earthworms ...	·00285	·02966	·03251

It may be inferred from these and similar experiments that earthworms have two effects in promoting fertility: (1) they act as cultivators of the soil, (2) they may produce plant food by decomposing organic matter more rapidly than could the micro-organisms of the soil. The cultivation effect is satisfactorily established. The proof of the direct production of plant food is less convincing; the experiments described below deal mainly with this question.

Repetition of the older work showed that marked crop increases could be obtained in pot experiments by putting earthworms into the soil:

Series 1.

	Experiment 1, 1907—1908		Experiment 2, 1907—1908		Experiment 3, 1909. Wheat. Dry matter, grams
	1st crop, Rye. Dry matter, grams	2nd crop, Spinach. Dry matter, grams	1st crop, Rye. Dry matter, grams	2nd crop, Spinach. Dry matter, grams	
Worms present	51·7	7·9	43·0	5·5	36·7
No worms present.....	36·7	6·2	34·1	2·8	30·3
Increase due to worms	15·0	1·7	8·9	2·7	6·4
Percentage increase ...	41	28	26	100	21

The pots contained 20 kilos of soil. In Expt. 1 30 worms were added; in Expt. 2, 10; in Expt. 3, 10.

The appearance of the plants showed that those in the pots containing worms were better supplied with nitrogenous food than the

others; they were darker green in colour, broader in the leaf, and contained a higher percentage of nitrogen in the dry matter.

The actual percentages were:

	Rye, Expt. 1	Rye, Expt. 2
Worms present	·993	·789
No worms present.....	·758	·689

The source of some of this additional nitrogen was indicated when, at the close of the experiment, the soil was tipped out and examined. No worms could be found. Casts and burrows were seen, but none of the worms survived to the end; they had died and their bodies had completely disintegrated. Analyses of a number of worms showed that they contained 1·5 to 2 per cent. of nitrogen in the fresh state and weighed about 0·5 to 0·7 grams each, the larger ones being rather the poorer in nitrogen; the worms used in these experiments contained about 10 milligrams of nitrogen each and therefore constituted a nitrogenous fertiliser. The following Table, setting forth the amount of nitrogen in the crops, shows that the additional nitrogen obtained from the soil containing earthworms is not very different from the amount of nitrogen supplied by the earthworms themselves.

	Experiment 1, 1907—1908		Experiment 2, 1907—1908	
	1st crop, Rye. Grams of nitrogen in crop	2nd crop, Spinach. Grams of nitrogen in crop	1st crop, Rye. Grams of nitrogen in crop	2nd crop, Spinach. Grams of nitrogen in crop
Worms present	·514	·135	·339	·093
No worms present	·279	·105	·232	·048
	·235	·030	·107	·045
Increase due to worms	·265		·152	
Approximate quantity of nitrogen in worms put into the pot	·3		·1	

In the second series of experiments the manurial effect of the worms was eliminated by putting into the control pots freshly killed worms equal in quantity to the living worms introduced into the experimental pots so that the added nitrogen was in each case the same. The pots were kept in the wire cage exposed to rain, and not, as in the previous

series, in the plant house; most of the living worms survived. The results were:

Series 2.

	Dry matter produced, grams		Percentage of nitrogen in dry matter		Weight of nitrogen in dry matter, grams	
	1st crop, Wheat (June—Oct. 1909)	2nd crop, Mustard (Oct. 1909—Feb. 1910)	1st crop, Wheat	2nd crop, Mustard	1st crop, Wheat	2nd crop, Mustard
Living worms added	12·9	3·2	1·168	2·425	·151	·078
Dead worms added...	10·1	3·1	1·454	2·694	·147	·083
					·004	·005
Increase due to living worms ...	2·8	·1	-·286	-·269	nil	
Percentage increase	27	nil	—	—	nil	

Each pot contained 10 kilos of soil and 10 earthworms.

Living worms cause an increase in yield, but over the two crops it is less than before. The significant difference from the earlier experiments, however, is that the living worms do *not* lead to any increased nitrogen assimilation; indeed there is actually a larger percentage of nitrogen in the plants supplied with *dead* worms. Thus it appears that the living worms have not contributed to the nitrogen food supply of the plant during the nine months of the experiment any more nitrogen than is contained in their own bodies—*i.e.* about 10 mgms. each—if indeed as much.

The soil was taken from an arable field in ordinary cultivation and was not especially rich in organic matter. In order to discover whether worms have more action in presence of plant residues, several pots were put up containing respectively mustard, vetches and grass, with and without worms. The results show a consistent increase in crop due to worms, but little beyond the error of experiment.

Series 3 *a*. March—Sept. 1908. Barley.

	Mustard dug in	Vetches dug in	
Worms present.....	3·88	4·55	grams of dry matter produced
No worms	3·38	3·88	" " "
Increase due to worms	·50	·67	" " "

Series 3 b. Arable soil, grass dug in (50 grams per pot).

	Dry matter produced, grams		Percentage of nitrogen in dry matter		Weight of nitrogen in dry matter, grams	
	1st crop, Wheat (June—Oct. 1909)	2nd crop, Mustard (Oct. 1909—Feb. 1910)	1st crop, Wheat	2nd crop, Mustard	1st crop, Wheat	2nd crop, Mustard
Living worms added	16·9	4·4	1·317	2·898	·222	·127
Dead worms added...	16·5	3·85	1·789	3·451	·295	·133
Increase due to living worms	·4	·55	− ·47	− ·55	− ·073	− ·006
					− ·079	
Percentage increase	2·4	14	—	—	−18	

Each pot contained 10 kilos of soil and 10 earthworms.

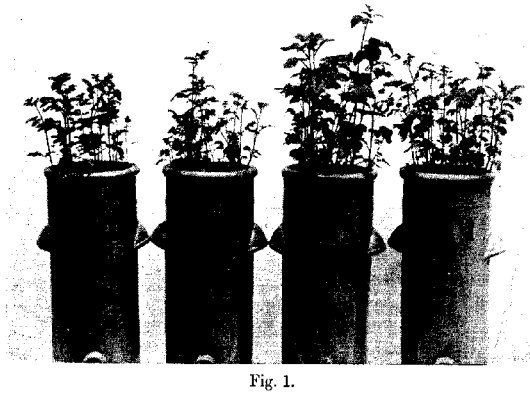


Fig. 1.

Series 3*b* is directly comparable with Series 2; the mustard crops are shown in Fig. 1. The effect of the added grass in increasing the crop and the percentage of nitrogen in the dry matter is very striking. The addition of dead worms to the pots has caused an increase of ·08 gram in the nitrogen content of the crop; the amount present in the worms themselves was about ·1 gram. Here, as before, there is no evidence that the living worms have in any way facilitated the decomposition of the organic matter or done anything in the way of producing

plant food that could not be done by the enzymes and micro-organisms already present in the grass or the soil.

Experiments made as part of the same series with a rich pasture soil led to the same conclusion. The crop supplied with dead worms was heavier, contained a larger percentage and larger total weight of nitrogen than the crop grown in pots containing living worms. The difference in weight of nitrogen amounts, as shown below, to ·095 gram, which is practically identical with the amount contained in the dead worms themselves.

Series 3*c*. Rich pasture soil: wheat (June–Oct. 1909).

	Dry matter, grams	Percentage of nitrogen in dry matter	Weight of nitrogen in dry matter, grams
Dead worms added	11·7	2·270	·266
Living worms added.....	9·2	1·862	·171
Increase where dead worms are present	2·5	·408	·095

In view of the special characteristics of partially sterilised soils already studied in this laboratory a series of experiments was made on soils treated with toluene. It was found that addition of dead worms caused a small increase in the crop and a larger increase in the percentages of nitrogen in the dry matter and in the total nitrogen taken by the plant from the soil. The weights of the crops were as follows:

Series 4. Toluened soil.

	Soil alone		Soil + grass	
	1st crop, Wheat. Dry matter, grams	2nd crop, Mustard. Dry matter, grams	1st crop, Wheat. Dry matter, grams	2nd crop, Mustard. Dry matter, grams
Living worms added.....	16·3	3·3	18·4	2·3
Dead worms added	16·8	3·9	16·8	7·0
Increase due to living worms	– ·5	– ·6	1·6	– 4·7

Each pot contained 10 kilos of soil and 10 earthworms.

As Series 2, 3*b*, 3*c* and 4 are strictly comparable the nitrogen statistics are here collected for convenience of reference.

Percentage of nitrogen in dry matter.

		Arable soil				Rich pasture soil
		Alone	+grass dug in	Toluened	Toluened +grass dug in	
Wheat.	Living worms...	1·168	1·317	1·395	1·690	1·862
	Dead worms ...	1·454	1·789	1·698	2·097	2·270
Mustard.	Living worms...	2·425	2·898	2·349	2·534	—
	Dead worms ...	2·694	3·451	2·057	2·548	—

Weight in grams of nitrogen taken up by crop.

		Arable soil				Rich pasture soil
		Alone	+grass dug in	Toluened	Toluened +grass dug in	
Wheat.	Living worms...	·151	·222	·228	·311	·171
	Dead worms ...	·147	·295	·285	·352	·266
Mustard.	Living worms...	·078	·127	·077	·058	—
	Dead worms ...	·083	·133	·080	·178	—
Excess in crops supplied with dead worms containing about ·1 gram of nitrogen		0	·079	·060	·161	·095

The higher percentage of nitrogen in plants associated with dead worms indicates that the general conditions are less favourable for plant growth than where living worms are present. It will be seen later on that there was a marked difference in the condition of the soil in the two cases, living worms having maintained an admirable tilth.

The effect of worms on the accumulation of nitrates in uncropped soils.

A parallel series of experiments was made in the laboratory to discover the rate at which nitrates accumulate in the soil under the influence of earthworms. Small pots each containing 3 kilos of arable soil, some containing living worms, others an equal quantity of freshly

killed worms, were kept in a room at about 10°—15° from November to the end of May, water being periodically added so that 15 to 18 per cent. should be present. There were double as many worms per kilo of soil as in the previous pot experiments. Examination of the soil was made at intervals, and samples were drawn for analysis.

The dead bodies rapidly became permeated with mould which spread into the soil forming a rather tough mass several times the size of the worm itself. After about three weeks, however, this had disappeared and no trace was left of the original body.

The amounts of nitrogen as nitrate found at different periods are given below in parts per million of dry soil:

	At beginning	After 10 days	After 27 days	After 74 days	After 6½ months	Increase in 74 days	After 6½ months	Excess in pots containing living worms after deducting nitrogen in dead worms m. grams of nitrogen	
								After 74 days	After 6½ months
A. Soil alone—									
Living worms	11·8	18·1	20·3	41·5	46·0	29·7	34·2	Nil	30
Dead worms	11·8	17·9	43·6	62·2	54·5	50·4	42·7		
B. Soil + 30 grams grass dug in —									
Living worms	11·8	25·2	42·7	66·6	87·5	54·8	75·7	79	142
Dead worms	11·8	21·5	41·5	57·6	56·7	45·8	44·9		
C. Soil + 30 grams grass mulch—									
Living worms	11·8	—	22·0	37·2	59·7	25·4	47·9	29	41
Dead worms	11·8	17·8	42·6	67·9	64·6	56·1	52·8		

6 worms were put into each pot and supplied approximately 60 milligrams of nitrogen.

In interpreting the above table it must be remembered that the dead bodies of the earthworms are undergoing decomposition; some of the nitrate in the pots containing dead worms therefore arises from this source. There is no means of finding exactly how much, but if we suppose that the whole of the earthworm nitrogen becomes nitrates and then deduct this from the total amount of nitrate actually found we obtain the nitrate produced from the soil by the action of bacteria in presence of dead earthworms. Deducting this again from the quantity of nitrate found in the pots containing living worms we obtain

the amount of nitrate produced through the activity of the living worms. These figures, expressed as milligrams of nitrogen, are given in the last two columns of the table; they are somewhat artificial because they assume that *all* the earthworm nitrogen has been nitrified and they would be lowered if this were not the case. But they are useful to us here because they represent the maximum amount of action that can possibly have gone on.

Dealing first with Set A, the pots containing soil alone and no added plant residues: for the first ten days no difference could be detected in the amount of nitrate formed, but between the tenth and the twenty-seventh day the dead bodies decomposed, bringing about a great increase in nitrate. In the subsequent period nitrates went on accumulating in both sets of pots, but later on they fell off in the pots containing dead worms. At the 74th day these pots contained 20 parts per million more than the others while at 6½ months the difference was only 8. Assuming that all the nitrogen in the dead worms was nitrified, the living worms have, in 6½ months, produced 30 milligrams of nitrate-nitrogen, which is about half the weight contained in their own bodies.

The rapid formation of nitrates from the corpses of worms confirms the high fertilising value shown in the pot experiments.

Hilderic Friend has recently (*Nature*, June 9th) raised the question whether worms and other living things in ooze contribute to its fertilising value. It seems certain that they do if, like earthworms, they contain as much as 1·5 to 2 per cent. of nitrogen.

The result of digging in grass (Set B) is very marked. It is clear that the living worms are increasing the stock of nitrates in the soil. After 27 days both sets of pots in this series contain equal amounts of nitrate, but from then onwards there is a steadily increasing difference. However, a detailed inspection of the living worms themselves showed that they increased in numbers, but the old ones died and of course gave rise to nitrates. Part, and apparently a large part, of the 142 milligrams of nitrate resulting from the addition of living worms is formed in this way.

These results appear at first sight to be inconsistent with the crop experiments already recorded but the circumstances are rather different. In the present series there are two worms and ten grams of grass per kilo of soil, twice as much as in the cropped soils and of course far beyond what would occur in nature. The conditions were purposely made highly favourable for earthworm activity. Yet even here, at the

very outside estimate and without allowing for the worms that died, the living worms have in six months only given rise to about twice as much nitrate as would be formed from the decay of their own bodies.

The third lot of pots (Set C) was designed to see how far worms would be effective in mixing a grass mulch in with soil. Comparison with Set A shows that there is not much result. At 27 days there is practically no difference between the two sets; at 74 days and at 6½ months the mulched soil containing dead worms is richer in nitrates presumably because of the smaller fluctuation in the water content, while the mulched soil containing living worms is at 74 days actually poorer, and at 6½ months not much richer, in nitrate in spite of the even water content and the grass that had been drawn into the burrows.

Determinations of ammonia by distillation with magnesia *in vacuo* showed, as usual, only about two parts per million of soil. This is not in agreement with Wollny's result quoted at the beginning of this paper where a great increase in ammonia was obtained, but I am satisfied that there must be some error in his determination as I have never been able to observe any accumulation of ammonia in soils where nitrification is going on. (See preceding paper.) It is conceivable that some fragments of decaying worms were present in his samples.

The experiments, like the pot experiments, show that earthworms do not play any great direct part in producing nitrates. It seems to matter but little whether earthworms are present or not so far as the conversion of plant residues and other organic materials into nitrate is concerned. In the most favourable circumstances, even during a long period, a worm was never responsible for an increase of more than 20, and probably not more than 10 milligrams of nitric nitrogen. To gain an idea of what this means in practice we may accept Hensen's estimate of 25,000 worms per acre in an arable field; these would at the best give rise in a season to about 250 grams of nitric nitrogen—rather more than half a pound, equivalent to 3¼ lbs. of nitrate of soda.

But if we are unable to allow earthworms any important part in the direct production of plant food we must acknowledge their power as cultivators. Fig. 2 shows the mass of soil tipped out from one of the pots; it had been penetrated in all directions by burrows so that drainage and aeration were both facilitated. Proof of their beneficial aeration effect was obtained by digging in early spring a piece of very wet ground so as to get the soil into a highly sticky condition. After three months the soil a little below the surface was greenish in colour,

indicating reduction; where, however, earthworms had been at work the surfaces of their burrows were red, proof that oxidation had taken place.

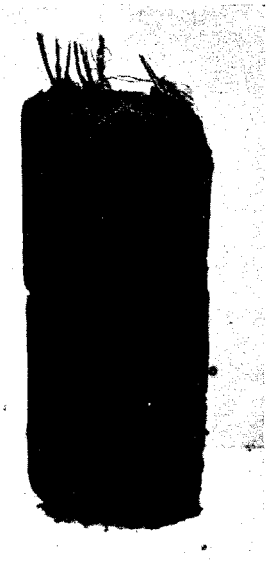


Fig. 2.

Their work on the surface of the soil is beautifully shown by its effect on algae and mosses; in pots containing no earthworms these forms soon appear on the surface of the soil, but not, however, where earthworms are present, since the surface is too often disturbed. Thus it is possible to pick out at once the pots containing earthworms by their freshly cultivated look in sharp contrast to the compact green surface when worms are absent.

Further, the cultivation effected by worms conserves the water supply. It was found that pots without worms required more water to keep them up to their proper degree of moistness than those containing worms.

In all these directions the soil conditions are made more favourable to plant growth and thus the plant is enabled to make fuller use of the food present. In the pot experiments the nitrogen assimilation where worms were active was associated with a higher production of

carbohydrate and therefore a lower percentage of nitrogen in the whole plant, than where the worms were dead. These effects are cumulative and may be even more important in the field than in pots.

Conclusions.

Earthworms do not appear to have any marked direct effect on the production of plant food. Organic matter seems to decompose with formation of nitrates equally quickly whether they are present or not.

They are rich in nitrogen, containing about 1·5 to 2 per cent., and they decompose rapidly and completely; thus they furnish a certain amount of plant food to the soil when they die.

Their chief work is to act as cultivators, loosening and mulching the soil, facilitating aeration and drainage by their burrows.

STUDIES OF THE CHANGES OCCURRING IN HEATED SOILS.

By SPENCER UMFREVILLE PICKERING, M.A., F.R.S.

In previous communications[1] it has been shown that soils heated to temperatures from 60° to 150° exhibit an inhibitory effect on the germination of seeds, due to the presence of some toxic substance, which must be a soluble organic, and, probably, nitrogenous, body, for the extent to which germination is retarded is roughly proportional, to both the soluble organic matter and the soluble nitrogen present. That so-called unheated soils, that is, soils which have not been heated above 20°—30°, contain some of this substance, was also probable from the fact that the results with unheated and heated soils all lie on continuous curves. It was shown, too, that treatment with antiseptics produced a chemical change in soils, closely similar to that produced by heating them to 60°—75°.

As a preliminary step towards obtaining some knowledge of the nature of this deleterious substance, a study was made of the changes which it undergoes when kept under various conditions: and this was preceded by an enquiry as to the trustworthiness of determinations of soil-extracts. Such determinations, if trustworthy, should have an extensive general application in soil analysis, for the soluble organic matter in soils, at any rate, in those of a similar nature, would probably afford a valuable measure of their fertility, and the determination of it would be much simpler than that of the nitrogen. From one series of determinations already published[2], as well as from others which have subsequently been made, it appears that the proportionality between the nitrogen and total organic matter in such extracts is generally very close.

The method adopted consisted of putting 100 grams of soil into a stoppered bottle with 1000 c.c. of water, shaking it up violently once

[1] *Journ. Agric. Sci.* II. 411, III. 33.

[2] *loc. cit.* p. 422.

every five minutes throughout two hours, leaving it to settle for 15 minutes, and then filtering through paper, an operation which generally occupied 8 to 12 hours. The filtrate was subsequently drawn through a Berkefeld filter, which was thoroughly cleaned by running several portions of the liquid through it first. 750 c.c. of the filtrate were then evaporated to a small bulk in porcelain, and transferred to platinum for evaporation to dryness. The residue, weighed after drying at 100°, and also after the strongest possible ignition over a gas blowpipe, was expressed as a percentage of soil dried at 100°. The preliminary filtering through paper can be dispensed with, though it facilitates the final filtration.

One obvious source of experimental error is the difficulty in weighing a large dish containing the hygroscopic residue. The error in determining the organic matter, which depends on two such weighings, cannot be placed at much less than a possible four milligrams. There are other sources of error which interfere with the absolute value of the results: the water retained after drying at 100° will be reckoned as so much organic matter, and the apparent weight of this will be further increased by the inorganic residue losing during the ignition any carbon dioxide and chlorides which may be present. On the other hand, volatile organic matter, if present, will be lost during the evaporation, and there may be a mechanical loss in the deflagration of the residue when much nitrate is present.

The compounds in the extracts are, doubtless, numerous, but three distinct features are noticeable. Some extracts, chiefly those of unheated and not very rich soils, leave a yellow granular deposit on evaporation, which, after ignition, is not easily removed by hydrochloric acid from the dish. In other cases a dark greasy scum separates before evaporation has proceeded far, and this adheres somewhat tenaciously to the porcelain. This is noticeable chiefly with very rich soils, and also with those which had been kept moist in closed vessels. In a third class, principally with soils heated to 125° and 150°, no solids separate on concentration, and the residue finally obtained forms a dark glassy mass. On ignition, this, and the residues from some of the less heated soils, especially from those kept moist in closed vessels, leaves an almost black deposit, which, however, contains no carbon, but dissolves readily in hydrochloric acid to form a deep brown solution, which, on heating, or on standing for some hours, becomes colourless or slightly yellow. This is probably due to the presence of manganese. The extracts, those of the last class especially, have a considerable action on platinum, the dish losing about 0·001 gram during each determination.

TABLE I. *Composition of soil extracts obtained under various conditions.*

	Conditions	Organic matter Per cent.	Organic matter Relative	Inorganic matter Per cent.	Inorganic matter Relative
	Time altered.				
1.	20 minutes	·0437	96	·0345	93
2.	40 ,,	·0470	102	·0365	99
3.	40 ,,	·0372	81	·0316	86
4.	80 ,,	·0438	97	·0400	108
5.	120 ,,	·0496	108	·0380	103
6.	160 ,,	·0410	90	·0395	107
7.	240 ,,	·0412	91	·0370	100
8.	320 ,,	·0512	112	·0417	113
9.	8 hours on filter	·0431	95	·0333	90
	Temperature altered.				
5.	6·8°	·0496	108	·0380*	103
10.	16·7°	·0467	102	·0427*	116
11.	23·5°	·0519	113	·0498*	135
	Proportions altered.				
12.	50 grams to 1 litre	·0462	101	·0520*	141
5.	100 ,, ,,	·0496	108	·0380*	103
13.	200 ,, ,,	·0323*	70	·0284*	77
	Mean, omitting those starred...	·0459	100	·0369	100

The results of the examination of the method of analysis, in which unmanured top soil from Harpenden was used, are collected in Table I, where the relative values entered are those obtained by comparison with the mean given at the bottom of the table. In the first nine entries the proportion used was 100 grams to one litre, at a temperature of 5°—10°, the time of digestion being varied from 20 to 320 minutes. As regards both the organic and inorganic matter, no effect whatever is produced by this variation, except a very doubtful increase when the time extended to 320 minutes. In No. 9, the soil was placed on a filter, and the litre of water was drawn through it, this occupying eight hours; but the results are sensibly the same as with the other method of procedure.

In the next three experiments the temperature, both of digestion and filtration, was altered, the proportions being 100 grams to the litre, and the time two hours. The organic matter dissolved does not seem to be sensibly affected by a difference of 16°, but the inorganic matter evidently is so, as it increases regularly with rise of temperature.

In the last three experiments the proportion of soil to water was altered. This does not sensibly affect the organic matter till the proportion exceeds 100 grams to the litre; but the amount of inorganic matter is affected, just as it was by temperature.

It is clear, therefore, that, especially as regards the dissolved organic matter, which is the point of chief importance, the method

gives perfectly satisfactory results, and these are not affected by such minor variations in the conditions as are likely to occur in practice. The variation of the results lead to a probable error of a single determination of the organic matter being 0·0028 per cent., equivalent to about ·002 gram, with the quantities taken, which is not greater than the probable weighing error.

In the investigations of the changes occurring in soils after being heated, the heating of the soil (which was Harpenden soil) was performed, as in former experiments, in closed vessels, two hours at the selected temperatures being allowed. The water present in the soil was 8·8 per cent., calculated on the sample after drying at 100°, its total water capacity being 33 per cent., or 50 parts of water to 100 of dry soil. Portions of 100 grams each of these soils were then kept under two different conditions: in one case, in open glass pans, with water to the saturation point added at intervals as the soil dried up, the soil, also, being broken up occasionally, as would have obtained in ordinary cultivation: in the other case, the soil, with water to the saturation point, was kept in hermetically sealed flasks, which, besides the soil, contained about 15 c.c. of air. In both cases the soil was kept in a table hothouse at about 15°. A few supplementary experiments were made with soil in flasks, kept in the hothouse, but in the dark, and also with soil kept at a winter temperature in the light. No precautions were taken in any case to prevent the reinoculation of the heated soils by bacteria, &c., but in no cases did any moulds appear on the soils during the experiments.

The water added in all cases was nearly that required for the saturation of the unheated soil, namely, 32 per cent.: but this is rather more than that necessary to saturate the soils which had been heated, for the total water capacity of the heated soils diminishes with the temperature of heating down to 27·4 per cent. for that heated to 150°. Some supplementary experiments, however, showed that the excess which would be present in these cases was without influence on the results: thus, soils which had been heated to 125° and 150° respectively, and of which the total water capacity was 30 and 27·4 per cent., respectively, were kept in sealed flasks for 10 days with 32·8 and 25·3 per cent. of water in each case; after which the alteration in the soluble organic matter in them was found to be

	With excess of water	With deficit of water
125°.....	+16 per cent.	+16 per cent.
150°.....	+ 9 „	+ 4 „

Although the heated soils can take up less water than the unheated ones, they retain it with considerably greater firmness. The weight of water added to 100 grams of the various soils in pans, to make up for the evaporation during 16 weeks, ranged from 400 c.c. in the case of the soils heated to below 100°, down to 314 c.c. with those heated to higher temperatures, though the loss did not altogether follow the temperature of heating, owing, no doubt, to irregularities in the air currents in the hothouse.

Sets of the soils were analysed after 10, 44 and 112 days, and other sets were used for germination experiments after 44 and 106 days. The original soil taken was also examined by analysis and germination to start with, and again after 119 days, it having been kept during that period in its original air-dried condition in stoppered bottles at a winter temperature. These bottles were capable of holding 30—40 times the quantity of soil present, and they were occasionally opened, so that there was ample opportunity for oxidation to occur, if any could occur under such conditions.

To ascertain the extent of the accidental variation which might be expected, duplicate experiments were made with soil which had been heated to 60° and to 125°, both in pans and in sealed flasks. The results were as follows:—

Extractive matter in soils after 62 *days.*

		Inorganic		Organic	
		Pans	Flasks	Pans	Flasks
60°.	A.	·0439	·0564	·0294	·0487
	B.	·0460	·0592	·0387	·0581
125°.	A.	·0593	·0938	·1182	·1967
	B.	·0503	·0831	·1038	·1883
Probable error of single experiment—		·0019	·0023	·0040	·0030
		0021		0035	

Thus, the probable error as regards the organic matter (·0035), is less than 50 per cent. greater than that determined above as the probable analytical error (·0028): as regards the inorganic matter the error is only ·0021. These errors are insignificant as compared with the total change in composition occurring during the time.

The results of the analyses are contained in Tables II, III and IV, and those of the germination experiments in Table V. In the latter, four sorts of seeds were used, and all the experiments were made in duplicate. The incubation period has alone been considered, the values entered being the average times required for germination, as compared with that required by the same seed in the unheated soil (*i.e.* heated

to 30°) in the same series. The rye used in the first series had, unfortunately, such a poor germination capacity, that the results with it were of no value.

TABLE II. *Soluble matter in soils after being kept watered for various lengths of time in open pans.*

After heating to	Percentage after				P.c. alteration in			Relative values after			
	Nil	10 d.	44 d.	112 d.	10 d.	44 d.	112 d.	Nil	10 d.	44 d.	112 d.
					Organic matter.						
30°	·0334	·0266	·0471	·0546	−20	+41	+63	100	100	100	100
60°	·0433	·0250	·0395	·0349	−42	− 9	−20	130	94	84	64
80°	·0684	·0342	·0498	·0377	−50	−27	−45	205	128	106	69
100°	·0967	·0705	·0723	·0592	−36	−25	−38	289	265	153	109
125°	·1763	·1362	·1178	·0727	−24	−33	−58	528	512	250	133
150°	·4187	·2688	·2333	·1242	−36	−44	−44	1254	1010	495	227
					Inorganic matter.						
30°	·0390	·0443	·0633	·0696	+14	+62	+78	100	100	100	100
60°	·0417	·0481	·0503	·0620	+15	+20	+49	107	109	79	89
80°	·0183	·0380	·0525	·0550	−21	+ 9	+10	124	86	83	76
100°	·0439	·0434	·0509	·0558	− 1	+16	+27	113	98	80	80
125°	·0674	·0451	·0553	·0495	−33	−18	−27	173	102	87	71
150°	·1125	·0716	·0747	·0640	−64	−34	−43	288	162	118	92

TABLE III. *Soluble matter in soils after being kept moist for various lengths of time in sealed flasks.*

After heating to	Percentage after				P.c. alteration in			Relative values after			
	Nil	10 d.	43 d.	116 d.	10 d.	43 d.	116 d.	Nil	10 d.	43 d.	116 d.
					Organic matter.						
30°	·0334	·0221	·0353	·0529	−34	+ 6	+ 59	100	100	100	100
60°	·0433	·0309	·0611	·0834	−29	+41	+ 93	130	140	173	158
80°	·0684	·0470	·0721	·0935	−32	+ 5	+ 37	205	213	204	177
100°	·0967	·0835	·1022	·1055	−14	+ 6	+ 9	289	378	289	200
125°	·1763	·1820	·1934	·1904	+ 3	+10	+ 8	528	824	548	360
150°	·4187	·4238	·4751	·4531	+ 1	+13	+ 8	1254	1918	1346	856
					Inorganic matter.						
30°	·0390	·0317	·0427	·0867	−19	+10	+123	100	100	100	100
60°	·0417	·0323	·0560	·0781	−23	+29	+ 87	107	102	131	90
80°	·0483	·0343	·0602	·0740	−29	+25	+ 53	124	106	141	85
100°	·0439	·0372	·0606	·0681	−15	+38	+ 55	113	117	142	79
125°	·0674	·0603	·0639	·0628	−11	− 5	− 7	173	190	150	72
150°	·1125	·1145	·1243	·1551	+ 2	+10	+ 38	288	361	292	179

TABLE IV. *Soluble matter in soils kept under various conditions.*

After heating to	Kept dry in bottles				Kept wet in sealed flasks					
	Original p.c.	After 116 days			For 43 days			For 116 days		
		P.c.	Alteration p.c.	Relative	In light	In dark	Diff.	In light	In dark	Diff.
					Organic matter.					
30°	·0334	·0228	− 32	100	·0353	·0347	− ·0006	·0529	·0606	+ ·0077
60°	·0433	·0283	− 35	124	—	—	—	—	—	—
80°	·0684	·0296	− 57	130	—	—	—	—	—	—
100°	·0976	·0558	− 42	245	·1022	·0902	− ·0040	·1055	·1074	+ ·0019
150°	·4187	·2598	− 38	1139	·4751	·4664	− ·0087	·1655	·1683	+ ·0028
					Inorganic matter.					
30°	·0390	·0431	+ 10	100	·0427	·0456	+ ·0029	·0867	·0591	− ·0276
60°	·0417	·0394	− 5	91	—	—	—	—	—	—
80°	·0483	·0381	− 21	90	—	—	—	—	—	—
100°	·0439	·0382	− 11	90	·0606	·0487	− ·0119	·0681	·0615	− ·0066
150°	·1125	·0554	− 51	129	·1243	·1335	+ ·0092	·1551	·1502	− ·0049
					For 62 days					
					At 15°	At 5°	Diff.			
					Organic matter.					
60°	—	—	—	—	·0667	·0534	− ·0133	—	—	—
125°	—	—	—	—	·1926	·1925	− ·0001	—	—	—
					Inorganic matter.					
60°	—	—	—	—	·0613	·0578	− ·0035	—	—	—
125°	—	—	—	—	·0637	·0884	+ ·0247	—	—	—

TABLE V. *Relative incubation periods of seeds in soils previously heated and kept under various conditions.*

Temp. of heating	Incubation periods						
	Wheat	Rye	Clover	Mustard	Mean		Mean B.
			1. *Soil taken, Oct. 19th.*				
30°	100	—	100	100	100		—
60°	148	—	50	137	116		—
80°	158	—	75	149	133		—
100°	152	—	106	117	120	143	—
125°	234	—	65	156	163		—
150°	220	—	69	190	186		—
			2. *Kept in pans till Dec. 2nd, 44 days.*				
30°	100	100	100	100	100		—
60°	109	106	115	122	113		—
80°	173	105	114	115	127		—
100°	148	129	123	108	127	121	—
125°	112	146	161	112	133		—
150°	165	165	190	147	167		—
			3. *Kept in pans till Feb. 2nd, 106 days.*				
30°	100	100	100	100	100		—
60°	85	105	119	104	104		—
80°	110	100	107	89	101		—
100°	93	120	154	121	122	107	—
125°	89	105	89	108	103		—
150°	97	100	102	122	105		—
			4. *Kept moist in flasks till Dec. 2nd, 44 days.*				
30°	100	100	100	100	100		114
60°	100	105	141	112	114		132
80°	110	128	83	148	117		130
100°	171	110	132	169	145	152	163 171
125°	214	123	100	250	172		189
150°	200	148	209	299	214		240
			5. *Kept moist in flasks till Feb. 2nd, 106 days.*				
30°	100	100	100	100	100		106
60°	105	112	116	147	120		127
80°	106	144	116	138	126		135
100°	88	180	123	124	129	139	140 151
125°	124	136	98	136	124		144
150°	123	183	258	223	197		207
			6. *Kept dry in bottles till Feb. 15th, 119 days.*				
30°	100	100	100	100	100		—
60°	80	100	88	86	91		—
80°	162	142	85	113	125		—
100°	134	154	77	143	120	122	—
125°	115	142	90	146	138		—
150°	179	141	95	123	134		—

TABLE VI. *Results obtained with soils kept moist in sealed flasks compared with those obtained from soils kept in open pans, represented by* 100.

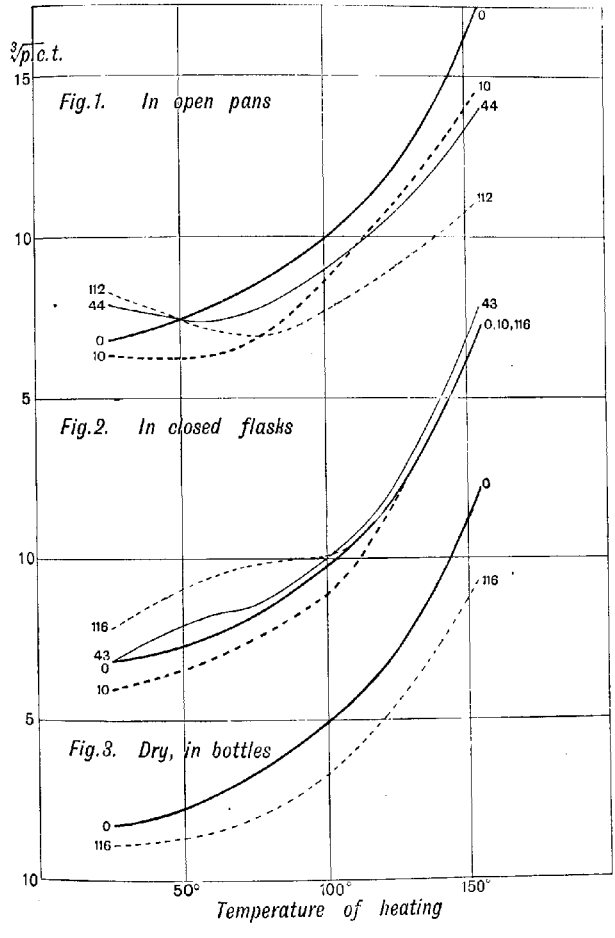
After heating to	Kept for 43—44 days			Kept for 106—116 days		
	Organic	Inorganic	Incubation	Organic	Inorganic	Incubation
30°	75	68	114	97	124	106
60°	155	111	117	242	126	122
80°	147	113	102	248	140	134
100°	141	119	128	178	122	115
125°	164	116	141	262	127	140
150°	204	166	144	365	242	197

Charts I and II represent the results of the determinations of soluble organic and inorganic matter, respectively, the cube root of the percentages of the former being taken for plotting, so as to reduce the steepness of the curves. The lettering at the ends of the curves gives the age in days of the soils, as dated from the starting of the experiments. A cursory glance at the figures is sufficient to show that changes of a very marked and complex character occur on keeping the soils, and that the alteration in the amount of soluble matter is sometimes negative and sometimes positive, and may extend to over 100 per cent. of that originally present. There are, however, certain well marked features in these changes. In the first place, the initial curves, marked 0, are in all cases very regular and simple. Looking at Figs. 1 in each chart, which apply to soils kept watered in open pans, the general result is (with one exception which will be dealt with immediately) that with the unheated, or less highly heated soils, the soluble matter, both organic and inorganic, increases as time passes, but with the more highly heated ones, the soluble matter diminishes, so that the curves representing the results of successive examinations cut each other at some point, this point varying from 50° to 80° as regards the organic matter, and from 80° to 105° as regards the inorganic matter; that is, soils which have been heated to these temperatures show little or no change in composition on being kept. This result is in full agreement with that of some preliminary experiments which have already been published[1].

[1] *Journ. of Agric. Soc.* III. 43. In these the point of intersection was at about 80°. Other determinations were made with a series of soils in which grass was grown for 72 days: the curves in that case were closely similar to those here shown, though not quite so regular, and the point of intersection was 75°.

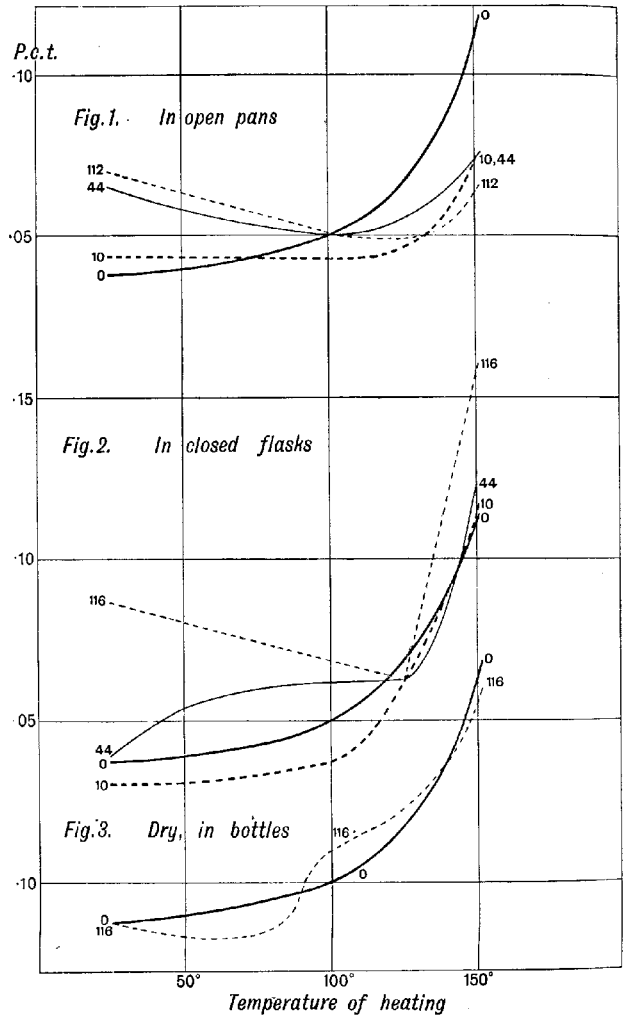
With the soils kept in sealed flasks (Figs. 2 on both Charts), there is a similar, though evidently more complicated, increase in the soluble matter in the case of the less heated soils, but, as regards the highly heated ones, there is no diminution, as in the other case, for the organic matter remains almost constant throughout, or, even increases

CHART I. Soluble organic matter in soils kept.



slightly, and the inorganic matter increases too, though only slightly so at first. It is clear, therefore, that the main changes occurring, or the

CHART II. Soluble inorganic matter in soils kept.



principal substances present, in the highly and less highly heated soils are not identical, and the results strongly indicate that, in these latter, the diminution of the soluble matter is probably due to oxidation, for oxidation would be at a maximum in the open pans, and at a minimum in the sealed flasks. The increase in the soluble matter with the less heated soils does not appear to be influenced in any definite manner by the access or absence of air.

The progress of the changes occurring is, however, not quite so simple, for, the increase in the soluble matter in the less highly heated soils is preceded during the first ten days by a decrease; this is shown equally by Figs. 1 and 2 in Chart I, and Fig. 2 in Chart II, whilst Fig. 1 in Chart II, without showing any such actual reduction, shows an irregularity tending in that direction.

These results indicate that the decrease in the amount of soluble matter is common to all the soils, whatever the temperature of previous heating, but that it is only in the case of the more highly heated ones, which are rich in this oxidisable matter, that the decrease continues throughout any considerable length of time; where less of it is present, as with the less highly heated soils, the supply soon becomes exhausted, and another change, resulting in an increase of soluble matter, becomes predominant. It is probable that this other change, just like the oxidation, occurs equally in all the soils, for it is noticeable that, as regards the inorganic matter, at any rate, an increase eventually asserts itself with the highly heated soils, especially when the conditions are unfavourable for oxidation: the experiments in closed flasks (Fig. 2, Chart II) clearly show this, and there is a tendency in the same direction, even in the experiments in open pans, for the final portions of the curve for 112 days (Fig. 1, Chart II) are high in comparison with those for ten and 44 days.

How far bacterial action may cooperate with atmospheric oxidation in reducing the amount of soluble matter in a heated soil when this is exposed to air is uncertain, but such a reduction does occur in the absence of all bacteria. Three samples of soil were heated in bottles to 127°: two of these were analysed and found to contain 0·7407 per cent. of soluble organic matter, whilst the third was kept in a very slow stream of dry air, under aseptic conditions, for eight weeks before analysis, and was then found to contain only 0·6478 per cent. The reduction in amount is not large, but the opportunities for oxidation would be very much more limited in this case, than in that of a sample of soil freely exposed to the air, and periodically moistened.

The soil was found to be perfectly sterile at the conclusion of the experiment.

Whether there would be any decrease in the soluble matter if air was entirely absent, cannot be settled by the present experiments, for the sealed flasks contained some air to start with, but it is most probable that there would be none in such a case.

That oxidation is the cause of the primary decrease in the soluble matter, is further borne out by the results with the dry soil kept in bottles for 116 days (Fig. 3, Charts I, II), where oxidation, though possible, would be less favoured than with soils kept moist. The results with the organic matter show a reduction in the 116 days closely similar to that shown in the other cases after ten days. As regards the inorganic matter (Chart II), the results are of too doubtful a character to afford any evidence in the matter.

That an air-dried soil, on being kept for a few months, should alter in composition, is a fact which the agricultural chemist will have to bear in mind when storing soil-samples. A similar change has been previously noticed in the case of soils treated with antiseptics[1].

The next step is to compare the results of the analyses with those of the germination of seeds. For this purpose the former must be treated somewhat differently. The absolute values for the incubation periods of seeds can only be compared together in cases where the experiments were made at the same time, for seeds of different ages behave differently: comparative values, taking the incubation period in the unheated soil as a standard, are alone available. The results of the soil analyses expressed in the same way are entered in the last columns of the Tables, and are depicted, together with the germination results, in Charts III and IV. The unlettered curves (- - - - - -) in Chart III give the results with the soil kept dry in bottles.

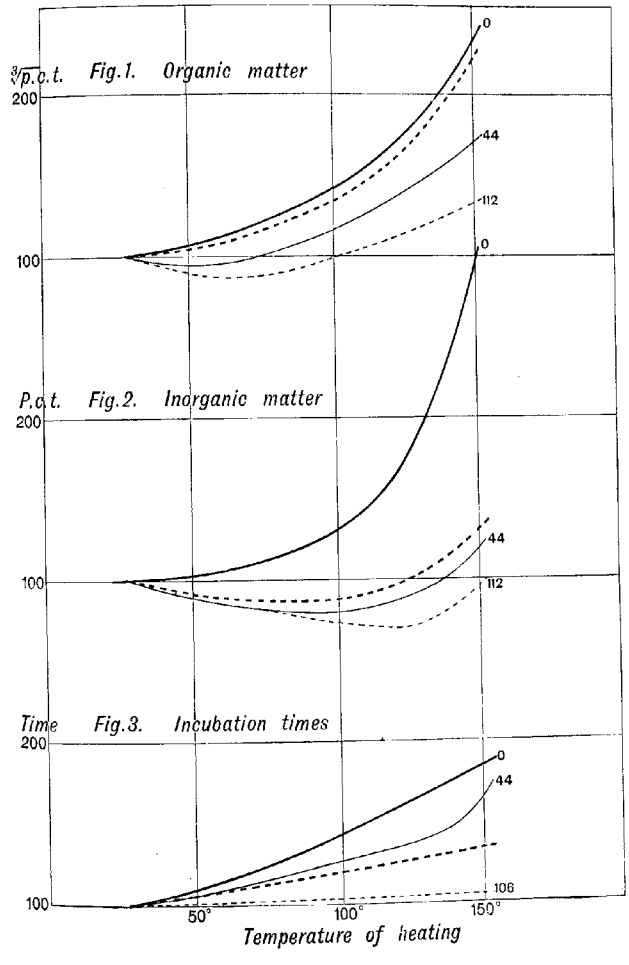
As regards the soils kept in pans (Chart III), it is evident that the diminution in the amount of both soluble organic matter and soluble inorganic matter in the more highly heated soils goes hand in hand with a diminution of their toxic action on the germination of seeds, the three curves for 0, 44 and 112 (or 106) days occupying the same relative positions in all cases. The soil kept dry in bottles does not behave in quite so regular a manner, the analyses placing it above the curve 44, and the germination results, below it.

Whilst it is evident that the soluble matter present in the highly heated soils is toxic towards germination, it would appear that that

[1] *loc. cit.* III. 35.

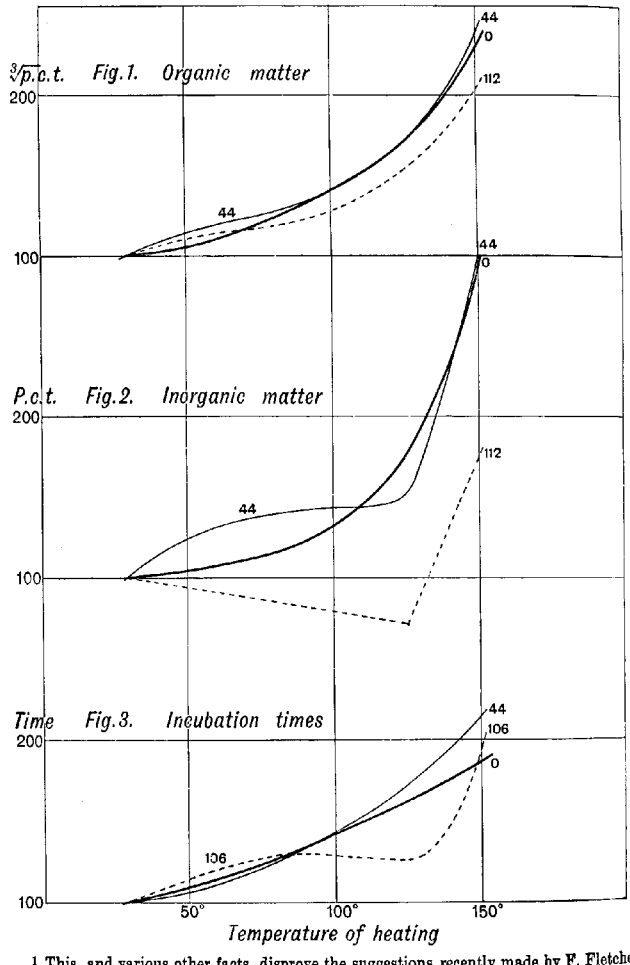
which is gradually formed in the less heated soils has no such toxic action: for with soils which have been kept for 44 and 112 days, the curves first dip (diminution of soluble matter) and then rise (increase in soluble matter), whereas the curves for the incubation periods rise

CHART III. Soluble matter in soils kept in pans, and incubation periods. Relative values.



throughout[1]. This would be seen better by taking the results at 100° as the standard of comparison; the incubation curves pass in a uniform

CHART IV. Soluble matter in soils kept in flasks, and incubation periods. Relative values.



[1] This, and various other facts, disprove the suggestions recently made by F. Fletcher (*The Cairo Scientific Journal*, No. 43, Vol. IV. April 1910) that delayed germination is due to a decreased rate of inhibition, consequent on the increased amount, and not on the nature, of the soluble matter in the heated soil.

direction through this point, whereas several of the analysis curves rise on both sides of it.

It is specially noteworthy that the toxic effect of the heated soils has nearly entirely disappeared by the 106th day, the curve (Fig. 3, Chart III) being almost an horizontal line, whereas the soluble matter in the various soils has by no means been reduced to uniformity: evidently the toxic substance is only one of the many substances formed when the soil is heated; it may be destroyed, and yet leave in the soil much of the extra soluble matter formed by the heating.

With the soils kept in closed flasks (Chart IV) there is a similar, though less exact, agreement between the analytical and germination results. The soluble organic matter in the soils heated to the highest temperature (Fig. 1), it is true, shows but little alteration with time, and the incubation periods (Fig. 3) do the same: but the three curves do not occupy quite the same relative position. It must be remembered, however, that the error of the germination results is considerable, and greater concordance could hardly be expected[1].

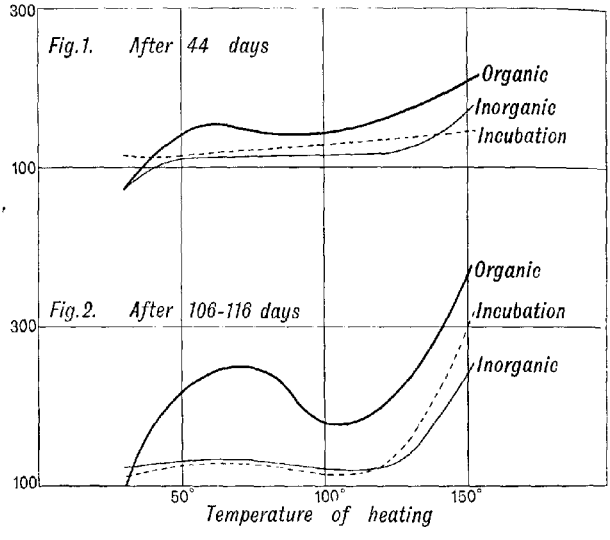
The incubation period for the soil heated to 125° shows a remarkable decrease on long keeping (106 days); and, on comparing this curve with those representing the soluble inorganic matter present (Fig. 2), a striking similarity between them is noticeable, showing that, in this case, the inorganic matter is a potent factor in the germination results.

The germination experiments with the soils kept in open pans and in sealed flasks having been made at the same time, these may be compared together: this has been done in Chart V, the curves "Incubation" being the values for the flasks when those for the pans are represented by a horizontal line at 100. Corresponding curves for the soluble matter have been drawn in the same way, the values for all of these being given in Table VI. The figures for the more extended period (Fig. 2) are of special interest in showing that it is the soluble matter in the highly heated soils which is the toxic substance, and not that which forms in the less heated soils on keeping, for the great rise in the soluble constituents from 100° to 150° is closely reproduced in the rise in the incubation period, whereas the great hump in the curve for the organic matter in the less heated soils produces only a very slight effect on the incubation curve.

[1] The germination curves have been smoothed more than the others: the results for soil heated to 100° lie a good deal off the curves 0 and 106, and, as may be seen from Table V, the values for this soil remained practically constant throughout.

Whilst the present experiments support in every way the conclusions previously drawn, that the toxic effect on germination is dependent on the increase in the amount of matter rendered soluble by heating the soil, the results do not show that direct proportionality between the increase in the incubation period and the increase in the

CHART V. Behaviour of soil kept in closed flasks compared with that kept in pans represented by 100.



soluble organic matter, which was previously observed. This may well be due to differences in the composition of the soil taken in the two cases, and, as a matter of fact, judging by the extracts, the two soils were very different (cf. Table II above with Table V on p. 422, Vol. II). Also, the amount of soluble inorganic matter seems to have a distinct influence on the incubation period in some of the present experiments, as has already been pointed out.

In connection with the most striking curve in Fig. 2, Chart IV, that for 112 days, it may be mentioned that the greasy scum which formed during the evaporation of the extracts in this case, was specially noticeable, both with the least heated and most heated soils, but was nearly absent with the intermediate ones, *i.e.* from those forming the

lowest part of the graph. A lesser amount of greasiness was observed with the soils kept in flasks for 44 days, but only with those which had been heated least, and none was observed with these soils when kept for only ten days, nor with any of the soils kept in pans. The greasiness, therefore, is evidently connected with the substances formed gradually by processes other than oxidation.

The effect of light on the changes occurring in soils kept moist in sealed flasks was examined in the experiments quoted in Table IV. The differences in the soluble organic matter are all negative after 43 days, and positive after 116 days: they are probably significant, but are too small for useful discussion. The differences in the soluble inorganic matter are mostly larger, but are not regular in character.

Some results with soils kept in closed flasks at an average temperature of 5°, instead of 15°, are entered in the lower part of the table—the values at 15° for 60 days are obtained by interpolation from the previously quoted results—but do not indicate any certain differences.

To ascertain whether there is any secular variation in the soluble matter in soils, a portion of ground in a cultivated orchard was protected from the weather by a glazed frame placed 18 inches above it, and samples of the soil were examined on about the tenth of each month throughout a year: at the same time some similar soil was kept in basins in an open shed, watered and stirred occasionally, and examined with the other. The results are given in Table VII. With

TABLE VII. *Soluble matter in soil on different dates.*

Date	Soil *in situ*		Soil kept in basins	
	Organic	Inorganic	Organic	Inorganic
Dec.	·0626	·0895	·0626	·0895
Jan.	·0483	·0587	·0665	·0952
Feb.	·0308	·0541	·0542	·1027
March ...	·0366	·0490	·0590	·0867
April	·0417	·0618	—	—
May	·0520	·0470	·0753	·0844
June	·0526	·0579	·0705	·0860
July	·0688	·0749	·0625	·0920
August ...	·0744	·0748	·0732	·0978
Sept.	·0789	·0820	—	—
Oct.	·0576	·0736	·0691	·1028
Nov.	·0690	·1102	·0559	·0728
Average...	·0561	·0695	·0649	·0910

the soil in basins there appears to be no variation in any definite direction, whilst, with that left *in situ*, there is a decrease, both in organic and inorganic matter, throughout the earlier part of the year and an increase subsequently: this, however, may be due merely to the rainfall acting through the exposed soil surrounding the protected patch.

SUMMARY.

The water extracts obtainable from soils are of constant composition as regards organic matter when the time allowed for the extraction varies from 20 to 320 minutes, the temperature from 7° to 23°, and the proportions from 5 to 10 of soil to 100 of water. The inorganic matter is not affected by the time, but is by the temperature and proportions.

The increase in soluble matter produced by heating a soil, and the accompanying toxic qualities towards the germination of seeds in it, is gradually reduced by exposing these soils in a moist condition to the air, even under aseptic conditions, but is not reduced, when the soils are kept moist in the absence of air. The destruction of the toxic substance is probably, therefore, due to oxidation.

Unheated soils, or soils heated only to a low temperature, exhibit on keeping an increase in soluble matter; this occurs whether air is admitted or not, and this change, therefore, is probably not an oxidation process: the substance formed, moreover, in such cases appears to have little or no toxic action on germination. This increase of soluble matter, due to the formation of a non-toxic substance, is preceded by a preliminary diminution of soluble matter, precisely similar to the diminution of toxic matter occurring continuously in the more highly heated soils: such toxic matter, therefore, appears to be present in all soils, whether heated or not, though, in the latter case, it is present in such small quantities that it soon becomes completely oxidised.

Air-dried soils, heated and unheated, when kept for some months show an appreciable reduction in soluble constituents, and also in toxic properties (where such properties were originally present), closely similar to the reduction exhibited by moist soils kept in air for about ten days.

PLANT-GROWTH IN HEATED SOILS.

By SPENCER UMFREVILLE PICKERING, M.A., F.R.S.

The existence of a toxic substance in heated soils, and the co-existence of two changes of an opposite character when such soils are kept—the one resulting in the oxidation and destruction of the toxin, the other in an increase of the soluble organic matter present—seem to offer an explanation of certain anomalies which have been observed in the growth of plants in these soils, to which allusion has already been made[1]; provided, always, that this toxic substance is toxic towards plant-growth in the same way as it has been found to be toxic towards seed-germination.

That plant-growth is generally more vigorous in soils which have been heated to about 100° than in unheated soils, is sufficiently proved by the work of Darbishire and Russell[2]; and the more recent work of Russell and Hutchinson[3] has shown that this increased vigour is mainly due to the altered bacterial conditions, though the increase in the soluble organic and nitrogenous constituents resulting directly from the heating must contribute, also, to the effect. The writer's experiments with apple trees[4] gave a further illustration of increased growth in heated soils, but, as intimated, more recent experiments, both with trees and with other plants, led to diametrically opposite results, growth being less vigorous in the heated soils, especially if the temperature of heating had been higher than 100°.

A summary of one of these series of experiments is given in Table I. The soil used was from the top spit of an unmanured plot of ground at Harpenden: it was heated to various temperatures for two hours in closed jars in an autoclave, the "unheated" soil being entered as having been heated to 30°. As the soil could not be prepared in large quantities,

[1] Pickering, *Journ. Agric. Sci.* Vol. III. p. 43.

[2] Darbishire and Russell, *ibid.* Vol. II. p. 303.

[3] Russell and Hutchinson, *ibid.* Vol. III. p. 111.

[4] Pickering, *ibid.* Vol. II. p. 434.

the weight taken in each experiment was rather less than 1 kilo, the plants being grown in glazed jars. They were not grown long enough to come to full maturity, the period of growth being from July to October; and no artificial heat was used. The number of plants in this series was considerable, and caused overcrowding.

TABLE I. *Crops grown in soils heated to different temperatures.*

Temp. of heating	Relative dry weights per plant											
	Verbena	Mustard	Spinach	Tomato	Clover	Lolium perenne	Festuca prat.	Festuca prat. H.	Mean		Mean incl. roots	
									Non-grasses	Grasses	Non-grasses	Grasses
I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII
30°	100	100	100	100	100	100	100	(100)	100	100	100	100
60°	76	35	140	84	71	199	105	(113)	81	150	82	200
80°	71	67	81	82	59	145	110	(100)	72	128	77	120
100°	43	44	99	89	71	168	105	(81)	69	137	76	164
125°	74	56	104	52	73	233	142	(74)	72	188	68	241
150°	52	45	89	73	61	329	158	(35)	64	244	60	244

In the case of the five plants other than grasses the results show a general decline with the increase in the temperature to which the soil had been heated, the mean results (cols. X, XI) showing considerable regularity, although various irregularities appear in the individual series. With the grasses, however, the variation is in the opposite direction, notably so with *Lolium perenne*. A second series with *Festuca pratensis* (H) is entered in the table: this was conducted with another sample of soil under somewhat different conditions, and the results differ from those of the first series in showing a decided decrease in crop with temperatures of heating above 100°. This series was made in duplicate, and in each case the results were closely concordant: they are not included in the means given in the table.

The values here entered are deduced from the weights of the dried crops, those deduced from the weights of the total plants (including roots) are given in cols. XII and XIII, and lead to substantially the same results, except in the case of *Festuca pratensis*, where the total plant weights give the effect of heating the soil as being uncertain, and, on the average, insignificant: the values in this case were

30°	*60°*	*80°*	*100°*	*125°*	*150°*
100	102	86	111	78	105

Another series of similar experiments, with a fresh sample of the same soil, was undertaken in the following season. The plants in each pot were thinned out to a number suitable to the size of the pots (from two with the tomatoes and tobacco, to forty with the grasses), and, after the plants had finished their growth, the soil was turned out, sifted from the roots, and a second crop of the same plants grown in the same portions of earth. The first crop was growing from April to August, and the second, from August to November, the plants being kept in a hothouse when necessary. The second crops were far from maturity when removed.

TABLE II. *Crops grown in soils heated to various temperatures.*

Temp. of heating	Relative dry weights per plant									
	Spinach	Tomato	Tobacco	Lolium perenne	Festuca prat.	Dactylis glom.	Mean		Mean incl. roots	
							Non-grasses	Grasses	Non-grasses	Grasses
I	II	III	IV	V	VI	VII	VIII	IX	X	XI
					First crops.					
30°	100	100	100	100	100	100	100	100	100	100
60°	185	151	248	209	155	252	195	205	193	243
80°	223	163	332	284	203	116	239	201	243	215
100°	247	155	339	320	343	323	247	329	248	329
125°	238	129	207	341	473	351	191	388	192	121
150°	230	94	176	500	561	353	167	438	120	371
100° D.	285	182	311	281	204	242	259	242	264	202
100° M.	262	193	328	340	394	307	261	347	260	282
					Second crops.					
30°	100	100	100	100	100	100	100	100	100	100
60°	135	—	180	98	87	89	158	90	128	109
80°	117	85	104	111	105	91	102	102	88	121
100°	128	86	125	129	188	113	113	143	100	167
125°	150	148	80	143	123	134	126	133	109	141
150°	172	200	68	155	164	141	147	153	122	142

The results are given in Table II. Looking at col. IX it will be seen that the action with the grasses is the same in this series as in the former case, there being an increase in vigour of growth with an increase in the temperature to which the soil was heated, and this effect is considerably more pronounced than in the former series, the growth in the heated soil reaching, in the extreme cases, to over five

times that in the unheated soil. The lowest figures in Plates XVI and XVII reproduce photographs of two of these grass crops.

With the non-grasses, col. VIII, the results show a marked difference from those in the former series, in that the growth in heated soils is in all cases greater, instead of less, than that in the unheated soil: they agree with them, however, in showing that a reduction of growth does occur when the temperature of heating is increased, though this reduction does not begin till beyond 100°, instead of being in evidence from the lowest temperature employed, as in the first series.

One feature which the numerical results do not show is that the deleterious effect of the highly heated soils diminishes with time, the plants showing little or no growth at first, but gradually recovering from the toxic action. This is evidenced by the photographs of tomatoes and tobacco on different dates[1], in Plates XVI and XVII, though not so fully as might have been, since no photographs were taken in the earlier stages, when the plants in the highly heated soils were so stunted that they did not show above the rims of the pots. With the grasses, no stunting was observed at any period of their growth, but it was not till growth was considerably advanced that the superiority of those in the highly heated soils became evident.

The disappearance of the toxic effect in the case of the non-grasses is further emphasised by the results obtained with the second crops, Table II and Plate XVIII: for with spinach and tomato such an effect has entirely disappeared, and these plants show a continuous increase of growth with higher temperatures of heating, just as the grasses do; it is only with tobacco that any signs of a toxic action remains. It will be noticed that the increase in growth due to the heating the soil is very much less with the second crops than with the first, the substance favourable to growth, as well as that which is toxic to growth, having largely disappeared, the soil reverting gradually to its original condition before heating.

In the first part of Table II are given two other experiments with soil heated to 100°; in one case, D, in an air bath with desiccation, in the other case, M, in an open jar in a current of steam, the heating in this case being kindly undertaken by Drs Russell and Hutchinson in the apparatus used by them in their work. The results obtained with these two samples and with the former sample, heated in a closed vessel in an autoclave, are very similar as regards the non-grasses, but, as

[1] The photographs have had to be reduced to different extents in different cases: the pots were all of the same size, and a two foot rule is shown in each figure.

regards the grasses, the soil heated with drying yields considerably lower growth values.

	Non-grasses	Grasses
Autoclave.....	247	329
Dried	259	242
In steam	261	347

			Soluble matter	
	Non-grasses	Grasses	Organic	Inorganic
Autoclave, 100°.....	247	329	·1034	·0772
Dried, 100°	259	242	·1410	·0829
In steam, 100°	261	347	·0739	·1037
Autoclave, 150°.....	—	—	·3664	·1027
Unheated	—	—	·0633	·0795

The soluble matter in these three samples is given above, and, as will be seen, shows considerable differences, but the connexion between these and the growth values is obscure. No doubt, the substances formed when the soil is heated under different conditions will not be the same, and the results cannot be expected to be proportional to the total soluble constituents. Data for two of the other soils used are also given above for comparison.

Besides the above experiments, a series was made with apple trees, the feature of these being that access of air to the soil was almost excluded. The trees taken were one-year-old trees on paradise stock, and these were grown in bottles containing half a gallon of soil, the tree stem passing through a waxed bung in which were fitted two tubes plugged with cotton wool. Water was introduced when necessary—as determined by weighing the bottles—through the tubes, but the amount of ventilation taking place through them would be very small. Four trees were grown for one season in soil heated to each of the temperatures selected, and two of these in each case were sterilised by immersion in mercuric chloride solution, it having been ascertained by preliminary trials that such immersion did not prevent, though it somewhat retarded, root-formation. The conditions were in all cases as aseptic as possible, but they would naturally be very imperfectly so. The bottles were buried up to their necks in the ground. Besides heated soil, soil treated with a mixture of carbon disulphide, a low-boiling paraffin oil and chloroform were used in one set.

The results previously obtained with apple trees when grown in large pots of soil freely exposed to the air[1] showed a considerable increase of vigour due to the heating of the soil: as measured by

[1] This *Journal*, II. 434.

the length of new wood formed—which agreed well with other features examined—the results were:

30°	*82°*	*200°* (? lower)
100	139	163

but in the present series the behaviour of the trees is in the opposite direction; the values with the individual trees showed considerable variations, but on taking the means, the growth measurements give a steady decrease with the increased temperature of heating of the soils, and the treatment with antiseptics gives results similar to those obtained by heating the soil to about 90°:

30°	*60°*	*80°*	*100°*	*125°*	*150°*	*Treated*
100	97	96	79	49	36	83

The weights of the new wood formed, however, show that shoots from the trees in the heated soil are somewhat stouter than those from trees in unheated soil, there being an increase in the weights formed up to about 90°, which was followed by a decrease:

30°	*60°*	*80°*	*100°*	*125°*	*150°*	*Treated*
100	108	112	90	80	46	106

The heated soils used in these experiments, it should be mentioned, contained less soluble matter than those used in the series with farm crops, for owing to the length of time necessary to prepare the experiments, and for the trees to start into growth, the soils could not come into play till some time after they had been heated. The soluble matter found in them in March, and subsequently in November, when the trees were removed, was:

	March		November[1]
	Organic	Inorganic	Organic
30°	·0768	·0845	·0916
60°	·0819	·0877	·0936
80°	·0915	·0877	·0971
100°	·1010	·0903	·1003
125°	·1469	·1070	·1137
150°	·2169	·1284	·1295
Treated	·0747	·1056	

The alteration occurring in the soils in these months is in accordance with that observed in the experiments described in the previous communication though, owing to the limited supply of air, they are smaller in extent.

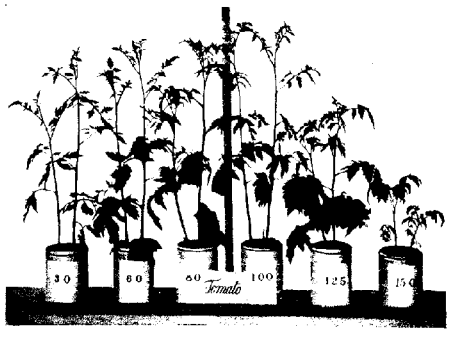
[1] The records of the inorganic matter have been lost.

A consideration of these various experiments makes it clear that, however contradictory the results may appear at first sight, they are fully in accord with the information obtained from a study of the changes occurring in heated soils, and with the experiments on the germination of seeds. On heating a soil, the soluble matter available for nutrition is increased, and changes in the bacterial condition are brought about, which—the latter especially—conduce to increased vigour of the plants growing in them: but the heating also results in the formation of some substance or substances which are actively toxic, and which tend to arrest growth. The proportion of toxin formed at low temperatures is small, and is generally insufficient to counteract those conditions favouring increased growth, but this proportion increases at a very rapid rate as the temperature of heating rises above 100°, and its baleful influence in such soils is generally the preponderating factor: hence the results obtained of increased vigour with soils heated up to about 100°, and of greatly decreased vigour with those heated to higher temperatures. But the toxic substance is unstable, and gradually disappears by the action of air and moisture, so that the results obtained in any individual series will vary considerably with the circumstances obtaining. When the soils are used at once after heating, and when the cultivation, and the access of air, are reduced to a minimum, the toxic action will prevail, and no increased vigour of growth may obtain in any case (*e.g.* the results in Table I, and those with apple trees in bottles); whereas, under conditions favouring oxidation, the toxic action disappears, and increased growth becomes the predominant feature: the gradual recovery of plants grown in strongly heated soils, and the smallness of the toxic action in the case of second crops, are illustrations in point.

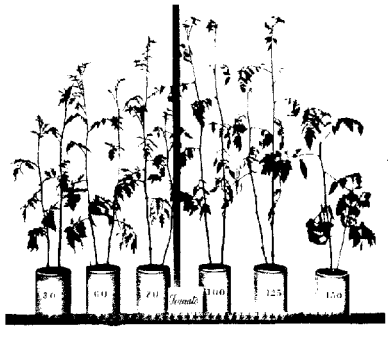
Whether the substance which is toxic towards plant-growth is actually the same as that which is toxic towards germination cannot be settled at present, but the heated soils appear to be equally toxic as regards these two processes, and the toxin in both cases is equally susceptible to oxidation: it is legitimate to assume provisionally that it is the same toxin which is active in both cases, and on such an assumption we can use the germination of seeds as a method of searching for its presence in soils. This is a point of importance, for germination experiments can be carried out in a few days, before a soil has had time to become altered, besides which, the germination of seeds is unaffected by such bacterial conditions and the supply of food as would mask any toxic action in the case of plant-growth.

Another point of great importance is the different susceptibility of different plants to the action of the toxin: it would be impossible, of course, to draw any conclusions from the present experiments that grasses generally are much less susceptible than other plants, but the results with grasses are sufficient to show that great differences in susceptibility exists. This may lead to some light being thrown on the obscure question of the action of grass on trees, and it also is suggestive of a fresh cause which may be assigned to the flourishing of certain species of plants in some soils and localities to the exclusion of others, in cases where differences of climate and food-supply seem insufficient to afford an explanation; for the formation of this toxin has been traced down to such a low temperature of heating, that it is impossible to avoid the conclusion that some of it must be present in so-called unheated soils.

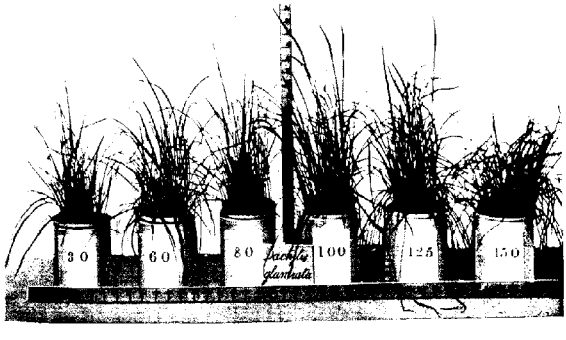
First crop.



Tomato, July 28.



Tomato, August 25.

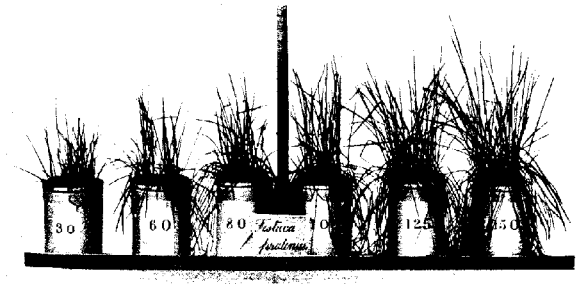


Dactylis glomerata, July 28.

First crop.

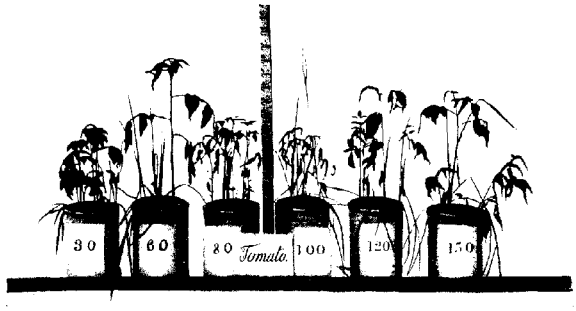
Tobacco, July 28.

Tobacco, August 25.

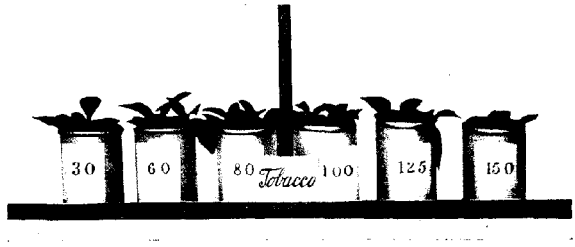


Festuca pratensis, July 28.

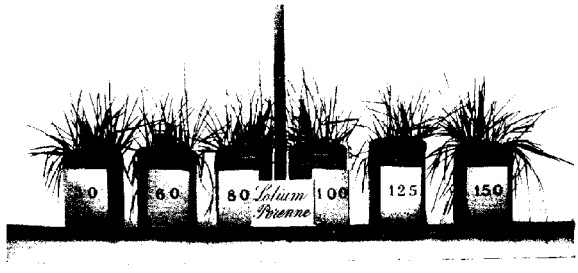
Second crops.



Tomato, November 26.



Tobacco, November 26.



Lolium perenne, November 26.

FIVE-DAY-SPRAYING.' THE BROWN TICK AND THE EAST COAST FEVER.

By W. F. COOPER, B.A.

In spite of the enormous mass of literature dealing with cattle-dipping, the actual amount of definite information on the subject is very small considering its vast economic importance. On this account this communication should be of interest.

At present, dipping is resorted to for two purposes. In different places, such as on the Borderland between Queensland and New South Wales, it is employed as a means of cleaning cattle in quarantine, so as to prevent the transference of the blue tick (the transmitter of 'Red-water' or 'Texas Fever') from areas infested with disease-transmitting ticks to areas not so infested. Again, dipping is resorted to in order to keep down the number of ticks on a farm to a reasonable limit. So far as I know, the dipping of cattle has never been found to clear any farm of ticks completely; but it is advocated by such men as Theiler, Lounsbury and others, more as a means of controlling the number of ticks on a farm, and keeping the pest in check, than with any definite object of getting rid of it entirely.

Dipping has been found to be of great value, because, unless the ticks are kept in check by some such method, they increase and multiply to such an enormous extent that grazing is practically impossible (Roberts 1899, p. 371). Such a condition of affairs had taken place at Gonubie Park, near East London, Cape Colony; where, though years before it had been renowned for its pasture land, yet a few years ago grazing was impracticable and unremunerative. After two years of consecutive fortnightly dipping, the ticks have become so much reduced that only a very small number are ever seen on any beast on the farm. This system of dipping the whole herd regularly every 14 days was advocated by the Cape Government: but it does not kill the brown tick, and it has been suggested, therefore, that one should dip every five days. The reason for this becomes clear, when the life histories of the different ticks are known.

The life cycle of a tick is as follows: from the egg emerges a larva, having three pairs of legs, no spiracles, and no genital pore: this gorges and then moults, giving rise to the nymph with four pairs of legs, a single pair of spiracles, but no genital pore: this nymph gorges, moults and gives rise to the adult, with four pairs of legs, a single pair of spiracles, and, of course, sexual organs.

The three chief ticks with which one has to deal—at least in South Africa—are the 'Blue Tick,' the 'Bont Tick,' and the 'Brown Tick.' The 'blue tick' remains attached to its host during its three stages, each of which occupies at least a week, or more if the weather is unsuitable; so that it is almost certain to be caught in one of its stages, by the fortnightly dipping. Therefore it is comparatively easy to reduce the number of *blue* ticks on the farm, and there is reason to believe that one might completely eradicate this pest from an estate in a few years. Experiments to determine this are being made at Gonubie Park.

The 'bont tick' drops off the host for each moult, and does not remain on the beast for more than about seven days, as has been shown by Lounsbury (1899), Dixon and Spreull (1898), and others: so that a fortnightly dipping may not catch this pest; since it is possible that, in one or other of its stages, it may get on a beast after the one dipping, gorge, and drop off before the next dipping. For this reason, it is not likely that the fortnightly dipping would completely clear a farm of the bont tick, except after a considerable number of years; it can very easily be kept in check by this means, however.

The 'brown tick' may remain on the host for no more than four days; so that there is a very great chance of the fortnightly dipping not catching this tick at all: it has been found, in actual practice too, that the system does not have a very great effect beyond that of checking this pest, and that it is a very difficult matter to get rid of it.

Concerning the blue and the bont tick, the necessity for getting rid of them is not so urgent, because the diseases of cattle transmitted by them are not of so serious a character[1]. The brown tick, however, is the

[1] To non-immune stock, Texas fever may be as fatal a disease as East Coast Fever. Smith and Kilborne (1893), *Annual Report, Bur. Animal Industries*, p. 274, give the mortality as 95%. Cattle may, however, be immunised against Texas fever by inoculation; also a herd which has been exposed to infection becomes naturally immune or 'salted.' As a matter of fact, nearly all of the S. African herds are infected with the disease in a latent form. It is not possible to obtain immunity in beasts against East Coast Fever, on the other hand, by any method tried up to the present, excepting in a few cases such as amongst the common native cattle in British East Africa, which do sometimes recover from an attack.

chief transmitting agent of East Coast Fever, a very fatal disease. The seriousness of this fever can be gathered from the fact that Bruce (1905) estimates the loss caused by it, in the Transvaal, at £200,000 in the year 1904; the number of cattle lost at 15,000. Though the disease exists at present in the Transvaal, it is chiefly confined to certain parts of Natal and Rhodesia: it also occurs in other countries such as British East Africa. There is great fear, however, that it may spread and cause a serious loss in parts at present free from it, and it is on this account that the problem of effective dipping and of the destruction of the brown tick, is so important.

It is impossible, here, to enter in detail into the matter of dipping and spraying: it will be sufficient to point out that it has been found that Sodium Arsenite is the only practical destructive agent for ticks: and that the maximum degree of dilution which is really effective against the blue tick was found by the Queensland Government to be one part of Arsenious Oxide in 500 parts of water (Brünnich, 1909). But solutions of this degree of concentration injure the skin of a beast if applied more than once in 12 days.

The Cape Government advocates the use of one part of the oxide in 300 parts of water (1 lb. to 30 gals.) used every 14 days: but this is to destroy the female bont tick (Hutcheon, 1905).

Watkins Pitchford, in 1909, published results which he had obtained with a mixture termed 'Laboratory Dip.' By the use of this mixture he was able to spray beasts every five days without injurious effect, and, with this treatment, he kept them free from ticks. In such work as he was doing, however, it was impossible to obtain normal conditions—those actually existing on the farm; and in my own work on dipping experiments at Gonubie Park, I have always found that, though it is most essential to make experiments in such a manner as those made by Watkins Pitchford, yet one cannot draw reliable conclusions from them as to what would occur in actual practice on the farm. For this reason the following results, which were obtained under normal grazing conditions, should be of interest.

One of the principal sources of error in working under laboratory conditions is that though one may examine a beast most carefully, yet it is excessively difficult to be certain that not one tick remains on it; one may be hidden, perhaps, by some fold near the scrotum, on the end of the tail, or in the hairs about the ear; also, although the brown tick is almost always to be found round the anus or about the ears, yet it is not invariably located on those areas.

Although Watkins Pitchford, throughout this most admirable piece of work, took the greatest possible care in the examination of his beasts, yet it must have been extremely difficult to be absolutely certain that not one tick was left. Such a matter is of the greatest importance, because one single tick is sufficient to transmit the disease.

I thought it would be of interest, therefore, to make some experiments on land where an outbreak of East Coast Fever was actually existent. By conducting the experiment under these conditions, one would have a practical test of the value of so dilute a dip applied every fifth day.

A point of interest is the fact that, in some work done by H. E. Laws and myself at Gonubie Park, it was proved that on the application of an arsenical solution to the skin of a beast, the arsenic penetrated the skin and was to be found in all the tissues of the beast; and further, that this took place very rapidly. Owing to the fact that the laboratory and the surrounding atmosphere were contaminated with arsenic, we were not able to obtain any definite data: but we were able to prove, by means of control experiments, that a considerable amount of arsenic was to be found in blood taken from the heart 18 hours after a fatal application of the arsenical solution, and 4 days after an ordinary dipping. Phillips, p. 474, mentions that arsenic given internally has been detected in the blood a few minutes after its application. It is known also, that certain organic compounds of arsenic, such as Atoxyl, Arsacetin, Soamin, etc., are useful in destroying certain blood parasites. It seemed to us possible, therefore, that even if the ticks themselves were not kept off the beast, yet by a regular dipping or spraying every fifth day (that is with an interval of four days between each dipping, as practised by Watkins Pitchford) the skin and blood might be impregnated with arsenic, to an extent sufficient to kill any micro-organisms transmitted to the beast by the tick. In this case, the presence of ticks on the beast would not be so important.

The opportunity of making an experiment to obtain information on these points occurred in British East Africa in July and August, 1909, and through the kindness and courtesy of Mr R. J. Stordy, F.R.C.V.S., Chief Veterinary Officer, I was able to carry out the experiments detailed below.

Six beasts were bought from the Government Farm near Lake Naivasha, British East Africa, a district known to be free from East Coast Fever: they were brought by rail to Nairobi, where they were immediately transferred from the trucks to the quarantine station.

Four of these beasts were sprayed immediately; two being left unsprayed, to act as controls. The day after spraying they were removed to a farm, the Kiambu Boma, where East Coast Fever was known to be prevalent and where beasts were proved by microscopical examination to be dying from it. These four beasts were sprayed regularly every fifth day with a solution of the following composition:

Arsenious Oxide (As_2O_3)	1 lb.
(dissolved in Sodium Carbonate (anhydrous) to form Sodium Arsenite)	½ lb.
Castor Oil	1 lb.
(made into soft soap with the correct amount of Potash)	
Castor Oil	1 lb.
(emulsified in the soap above)	
Water to 700 lbs. the volume of which is	78 galls.

Watkins Pitchford's formula (p. 23) contained eight and a half pounds of Arsenite of Soda in 400 gallons of water. We were able to show most conclusively, at Gonubie Park, that the 'strength' of a dip is determined by the Arsenious Oxide contained in it and not by the amount of Sodium Carbonate present. Spraying with a 2 °/₀ solution of Sodium Carbonate had no ill effect whatever; whereas Arsenious Oxide dissolved in Glycerine had the same effect as the same amount of Arsenious Oxide dissolved in Sodium Carbonate, both being used at the same dilution. Unfortunately, there was no statement as to the percentage of Arsenious Oxide in the Arsenite of Soda used by Watkins Pitchford, and it has been found difficult to obtain Arsenite of Soda of known and constant composition. One may assume, however, that it would contain 66 °/₀; so that the eight and a half pounds would contain 5·61 lbs. of Arsenious Oxide: this in 4,000 lbs. of water (*i.e.* 400 gallons) gives a solution of 1 part of Arsenious Oxide in 713 parts of water. My spray fluid, therefore, contained about the same amount of Arsenious Oxide as the 'Laboratory' spray fluid.

As to the quantity of soap and oil for the emulsion: it had been shown, by Lounsbury and also by experiments carried out by us at East London, that the value of any soap and any oil was due to the physical properties of such *as an emulsion*: that paraffin oil emulsion at a strength of 50 °/₀, as Watkins Pitchford used it, has absolutely no killing power when used by itself and that its value in a dip is due to its physical properties: that castor oil and castor soap gave a more efficient emulsion than one of paraffin oil: and that the most efficient quantity for castor oil and castor soap appeared to be about equal in amount to that of the Arsenic Oxide. In these two points my fluid differed from that of Watkins Pitchford: but I believe from earlier work that my

proportion of oil and soap, and the use of castor oil in place of paraffin oil, ought to give results superior to those of Watkins Pitchford. Not only that, but Watkins Pitchford's results were not available at the time, and I could not know what quantities of paraffin oil he used. Therefore I used a mixture which I had tried before and had found to be efficient.

Unfortunately I was unable to carry out all the experiments myself, and Mr Stordy very kindly placed Mr Ghulam Hassan, a qualified Veterinary Surgeon, at my disposal. Mr Hassan, therefore, did the regular sprayings and kept full notes: blood smears and spleen punctures were made by him when necessary and were examined by Dr P. H. Ross of the Government Bacteriological Laboratory at Nairobi. The following are the results:

Six beasts used; designated by letters *A* to *F*. All beasts were half bred between the native and imported English stock: all were bullocks, and varied in age from 10 to 16 months. The fluid was sprayed on to a beast with a Deeming 'Success' Pump and Vermorel nozzle.

Beasts A and B were left unsprayed, being kept under the same conditions as the others, on the same land, enclosed at night in the same kraals.

Beast B was sprayed by myself once, on July 19th, immediately after removal from the truck: but he was too wild to be sprayed at the farm, and it was necessary to use him as a control. So that he was sprayed once only. *Beast A* was never sprayed.

Beasts C, D, E and F, were sprayed as recorded.

Symptoms.

Note 1. *Beast F.* Aug. 13th.

Hidebound. Eyelids swollen. Eyes irritable and watery. Diarrhoea and paraplegia: unable to stand. Thinking that it was suffering from arsenical poisoning, it was left: it died the same night.

P.M. Examination. Stomach and intestines were slightly congested and spleen was enlarged. All other organs were found in good condition. Spleen smears gave negative results.

Note 2. *Beast A* (Control). Aug. 18th.

Off food. Watery discharge from eyes and nose. Prescapular, prefemural, and parotid glands were much swollen. Diarrhoea. Temp.

106·0° F. Blood smears showed rings and rods present and numerous. Died Aug. 21st. *P.M.* Spleen smears showed *Pyroplasma parvum.*

Note 3. *Beast B* (Control). Aug. 18th.

Hidebound, appetite good. Constipated. Temp. 106·4° F. Blood smears showed rings and rods present and numerous. Could not be examined on Aug. 23rd, because too wild to hold.

Aug. 28th. Temp. 104·0° F. Spleen punctures showed poikilocytosis: polychromatophilia: very scanty *P. bigeminum*; scanty rings and no blue bodies in the spleen.

Sept. 1st. Was lying about 100 yds. from kraal, unable to stand.

Note 4. *Beast E.* Aug. 28th.

Temp. 107·0° F. Spleen punctures taken showed blue bodies; rings and rods present and numerous. Two *P. bigeminum.*

Died on Aug. 29th.

Note 5. *Beast D.* Sept. 1st.

P.M. Examination. Froth was issuing from the nostrils. On opening the abdomen the spleen was found to be of normal size: lesions on both the kidneys: ulceration of the fourth stomach: and enlargement of the liver. Lungs were congested and the air-passages were full of foam. Yellow coloured fluid was found in the pericardium.

Spleen smears showed rings and rods. Blue bodies present and numerous.

Results.

One of the beasts, *F*, died from arsenical poisoning, certainly not from East Coast Fever.

The two controls *A* and *B* both suffered from East Coast Fever. *A* died, *B* remained in bad condition but parasites disappeared at the time of the last spraying. Amongst the common native cattle (the experimental beasts were half bred) recovery from East Coast Fever is not uncommon: beast *B* certainly suffered from East Coast Fever, and it may be assumed that the recovery was due to the influence of native blood.

At the same time that the controls were suffering from East Coast Fever, the four beasts sprayed were quite well: one remained well for 5 days more, two for 12 days longer.

Spraying	Date and Time	Beast A (Control)	Beast B (Control)	Beast C	Beast D	Beast E	Beast F
1st	July 19th, 6.6 p.m.	——	——	——	——	——	——
2nd	July 24th, 6.0 a.m.	Quite well.	Quite well.	Quite well.	Quite well.	Quite well.	Quite well.
3rd ..... Spray used more dilute, 1 : 800 in this and subsequent sprayings.	July 29th, 5.30 p.m.	Quite well.	Quite well.	Quite well.	Skin slightly blistered on perinaeum, inside thighs and on dewlap.	Quite well.	Skin slightly inflamed, cracked on hairless parts.
4th	August 3rd, 5.30 p.m.	Beast in better condition. Live ticks found on all.	Quite well.	Quite well.	Quite well.	Quite well.	Quite well.
5th	August 8th, 6.0 a.m.	Quite well.	Quite well.	Covered with ticks. None dead.	Quite well.	Quite well.	Better. Cracks dried. All ticks dead.
6th	August 13th, 4.0 p.m.	Quite well.	Quite well.	Progressing. Covered with ticks.	Quite well.	Quite well.	Very ill. Died in the night. Arsenical poisoning. (Note 1.)
7th	August 18th, 4.0 p.m.	Ill. Suffering from East Coast Fever. Died on Aug. 21st. (Note 2.)	Ill. Skin inflamed. Suffering from East Coast Fever. (Note 3.)	In good condition. Covered with ticks. Skin somewhat inflamed.	Quite well.	Quite well.	——

8th	August 23rd, 11.0 a.m.	——	Not well. Could not take puncture: too wild to hold.	In better condition.	Quite well.	Quite well.	——
9th	August 28th, 10.30 a.m.	——	Ill. Temp. 104·0° F. East Coast Fever not shown bacteriologically.	In good condition. Temp. normal.	Quite well.	Ill. East Coast Fever. (Note 4.)	——
	August 29th	——	——	——	——	Dead from East Coast Fever. Spleen showed numerous blue bodies.	——
	August 30th	——	Ill. Temp. 106·0° F. Blood contained very scanty *P. bigeminum*.	Ill. Temp. 106·0° F. East Coast Fever. Rings and blue bodies plentiful.	Ill. Temp. 107·4° F. East Coast Fever. Rings and blue bodies plentiful.	——	——
10th.....	September 1st, 7.0 a.m.	——	Lying abont 100 yds. from hut, unable to stand.	Very ill. Temp. 107·0° F. East Coast Fever. Blood showed rings and rods, also *P. bigeminum*.	Found dead. East Coast Fever. (Note 5.)	——	——

C was not dead on Sept. 1st, but was very ill and suffering from East Coast Fever.

D died on Aug. 31st of East Coast Fever.

E died on Aug. 29th of East Coast Fever.

F died on Aug. 14th of arsenical poisoning.

Conclusion.

The Dip. I was astonished to note that the dip with Arsenious Oxide at 1 : 700, and later 1 : 800, had such a severe action on the beasts. It must be remembered, however, that they had not been sprayed before: this is a factor of considerable importance. But it is my opinion that the severity of the dip was to be accounted for by climatic conditions: for, in the work done in the Cape Colony, many reasons led one to conclude that this has a considerable effect. Lounsbury (1902, p. 10) and also Hutcheon (1905, p. 534) made similar observations when using paraffin.

The effect of the Dip. It is obvious that the spray was as strong as one could employ it. It is equally certain that this did not keep off the brown ticks.

The period at which beasts were noted to be ailing, and at which microscopical examination showed that they were suffering from Fever, was certainly delayed to some extent: but whether this was a mere accident, or was on account of arsenic in the system, must be left to the reader to decide; it would appear to me that the latter is the case.

It must be pointed out that brown ticks were *observed* to be on the beast on July 29, and if this is the date on which the ticks first got on the beast then Aug. 28th would be just about the time when they would show symptoms. But to one who knows how the ticks infest every blade of grass, almost, it would not seem to be possible that the beasts were not infested immediately upon their arrival at the farm: in this case, they should have shown symptoms on Aug. 18th and should have been dead in a day or two. This actually happened in the case of the two control beasts, the one dying on Aug. 21st, and it is practically certain that the others would be infested at the same time.

As to whether a 'five-day-spray' will eradicate the brown tick from a farm, these experiments give no indication. For this end, the experiments would have to be continued over many months: and, in that case, one has to take into consideration, the enormous amount of labour when

dealing with a herd of any size; the cost, not only of the dip, but of the labour; the fact that the cattle must be near the farm; and, finally, the effect of the arsenic on beasts.

Though these trials are the first of their class to be recorded, and only form a preliminary experiment, I certainly do think that they show that, until we have more experimental data, very little reliance can be put on the 'five-day-spraying' as a *preventive* against East Coast Fever; at any rate, unless the cattle have been dipped continually for some time previously. Until further work has been done with spraying, on a farm actually infected with East Coast Fever, dealing both with cattle used to spraying, and also with cattle quite unaccustomed to it, I think that it would be extremely unwise for the Cape Government or any other body to rely on the 'five-day-spraying' as a means of preventing it from spreading to so important a district as the Transkei Territories[1]. It seems that one would have to recur to the stamping out method, at present in vogue, when any outbreak actually occurs: yet, at the same time it would appear that this system of dipping every fifth day is of considerable value in delaying infection.

In Watkins Pitchford's work, p. 23, however, it is to be noted that the dip was not effective until after the tenth spraying. *It may be that, in this experiment, the beasts ought to have been sprayed* 10 *or* 15 *times before being exposed to infection.* Perhaps, had that been done, the experiments would have been more successful; and, in that case, the method could be relied upon as a quarantine method to replace the drastic measure of stamping out. This, also, would account for the failure of the trial, of which the results are recorded here. Further experiments are to be made in British East Africa, in which the beasts will be placed on high land not infested with ticks; they will be sprayed twenty times, and then placed on land—together with controls—where East Coast Fever actually occurs. From this, we hope to learn whether the beasts can accumulate sufficient arsenic to be able to resist, either the infestation of ticks or the infection of the disease transmitted by them. That such an accumulation may occur is borne out by the experiments of Gubler, in which the elimination of arsenic in the urine ceased after six weeks from the last dose; but on the administration of Potassium Iodide a further quantity was eliminated

[1] The Transkei Territory is that lying north of the Kei river; between it and the southern border of Natal. East Coast Fever occurs right on the Natal border and most stringent regulations and most careful watch are observed on the border line.

Since these observations were made East Coast Fever has crossed the border into the Transkei in spite of stringent regulations and precautions to prevent the spread of infected ticks.

(Phillips, 1904, pp. 474 and 475). It is hoped that within a few months sufficient information will be at hand to enable others to make this method thoroughly reliable.

The most serious drawback to a more extended and more thorough trial of this 'five-day-spray' is the lack of funds. But the matter is one of such urgent importance to the Transkei, and indeed to all districts, that the Governments interested in it ought to take it upon themselves and to depute Watkins Pitchford to continue his experiments where East Coast Fever actually exists: unless, of course, some public-spirited person will come forward and assist in preventing the spread of so serious a disease.

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A METHOD FOR THE STUDY OF SOIL FERTILITY PROBLEMS.

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SEVERAL years ago the writer's attention was called to the apparently favourable influence of field peas on oats when the two were grown together. The rank growth of the oats, their dark-green colour and the delayed ripening gave every indication of an abundant supply of available nitrogen compounds. On the other hand, oats seeded without the peas, at about the same time, were less rank in their growth and matured at an earlier date. Further observation and inquiry strengthened the impression in the writer's mind that the associative growth of legumes and non-legumes is frequently advantageous to the latter in that they are supplied with nitrogen compounds derived either from the decay of the fibrous roots of the legumes, or from the soluble materials passing out of the roots into the surrounding soil.

Because of the extreme scientific and practical interest attached to this question, it was decided to submit it to a careful study. This seemed the more desirable in view of the very frequent growing of mixtures of legumes and non-legumes not only in dairy sections, but also in those where the raising of cattle, swine and sheep, and the growing of general farm crops are prevalent. In North America winter vetch is grown together with rye and wheat, and in more favoured locations with barley and oats. Similarly, crimson clover is occasionally grown together with barley or oats for forage purposes. Cow peas and soy beans are commonly grown together with corn, sorghum, Kafir corn or millet, and are either harvested as green forage, or are ensilaged or "hogged off." Timothy and other grasses are frequently mixed with

alsike, red or mammoth clover; while in natural pastures the clovers and other legumes are more or less prominent. In our Southern States corn and lima beans are grown together in order to provide the latter with proper support. Other mixtures of legumes and non-legumes have also been grown in America and Europe.

When considered from the nitrogen standpoint these combinations of legumes and non-legumes reveal possibilities of great economic importance. Should it be demonstrated that non-legumes could be provided with an abundant supply of nitrogen even in poor soils, by being grown together with legumes under proper conditions, it would become practicable not only to dispense with all or a portion of the nitrogenous manures employed for certain crops, but also to secure non-legumes with an increased proportion of protein in the dry matter. In accordance with this thought a method was devised for the study of the reciprocal effects of legumes and non-legumes.

The method itself is very simple, as is shown in the figure. The outer vessel is an ordinary five-gallon glazed earthenware pot. The inner and smaller pots are made out of a very porous flint mixture, and differ only in that the pot on the left is glazed, while the pot on the right is unglazed.

In arranging the experiment for the study of the relations of legumes and non-legumes, the small and large pots were filled with white quartz sand, the smaller pots being placed inside the larger.

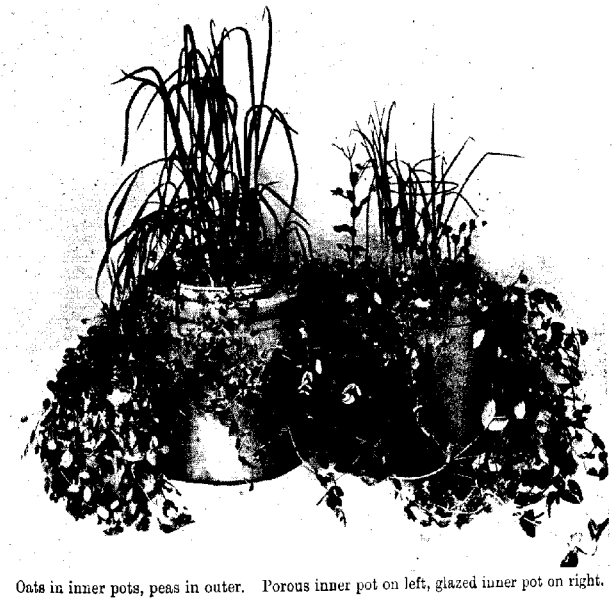
There were secured in this manner two portions of soil identical in composition and supplied with the same fertilizer materials. The two portions of soil were separated from one another by a porous wall in the one instance, and by a non-porous wall in the other. The legumes were planted in the large pot, that is, in the outer portion, while the non-legumes were planted in the small inner pot. Previous to planting the sand used as soil was supplied with all the essential mineral constituents and with a small amount of soil infusion in order to supply the bacteria for the inoculation of the legumes.

It was reasoned that if the legumes allow soluble nitrogen compounds to pass out of the tubercles and the roots, these soluble compounds will diffuse through the porous wall of the unglazed pot and supply nitrogen to the non-leguminous vegetation in the inner pot. On the other hand such diffusion cannot take place through the walls of the glazed pot and the non-legumes growing in it will starve for lack of nitrogen if none is supplied in the fertilizer material.

Accordingly field peas and oats were planted in the outer and inner

pots respectively. Pure quartz sand was used as soil, and was supplied with all the essential mineral constituents. No nitrogen was applied, and the plants had no other source of supply except the atmosphere, or the slight amounts of nitrogen present in the seed or in the water used. The outer pot contained about 80 lbs. of sand, the inner pot about 20 lbs. The moisture conditions were kept uniform.

The figure shows the glazed inner pot and corresponding large pot on the right and the unglazed inner pot and corresponding large pot on the left. It will be noted that the oats in the unglazed pot were



Oats in inner pots, peas in outer. Porous inner pot on left, glazed inner pot on right.

sturdier and were making better growth. The photograph does not bring out the much deeper colour of the oats growing in the unglazed pot. There was no doubt, however, that at the time this photograph was taken, that is, when the plants were about 10 weeks old, the oats in the unglazed pot were securing nitrogen from some source that was not available to the oats in the corresponding glazed pot. Every indication was thus supplied that soluble nitrogen compounds were diffusing through the unglazed porous wall and were being utilized by the oats.

We have here a striking proof of the ability of oats to secure an adequate supply of nitrogen when growing together with field peas in a soil devoid of nitrogen. We have proof, further, that the nitrogen compounds supplied to the oats were soluble, and diffusible through porous earthenware. Subsequent weighing and analysis of the oats grown in glazed and unglazed pots, respectively, showed not only a much larger yield of dry matter and of nitrogen in the latter, but of dry matter containing nearly double the proportion of nitrogen as compared with that grown in the glazed pots. Since this is only a preliminary paper the analyses and other data are not given. Moreover, the purpose of this publication is not to discuss our results on the associative growth of legumes and non-legumes (we reserve these for publication at a future date), but to point out the wide usefulness of the method employed for the investigation of a great variety of soil problems.

The method may be employed, as we have employed it, for the study of the influence of various crops on the bacterial flora of soils. For this purpose the crops may be grown in the outer pots and the soil in the inner pots, glazed and unglazed, may be left uncropped. The samples drawn from the inner pots may be examined bacteriologically, and the differences found in the glazed and unglazed pots may be ascribed to the influence of the crop. Similarly the method may be employed for the study of the effect of various fertilizers on certain groups of soil bacteria; for the study of the influence of different crops on one another when grown continuously and in rotation; and for the study of the so-called toxic effect of plant-root excretions.

In order to secure satisfactory results with this method it is necessary to have special porous mixtures for the preparation of the inner pots. We have been able to secure satisfactory porous pots by mixing the clay with 25 per cent. of hard coal and 25 per cent. of soft coal. The pots made out of this mixture are fired in the usual way. As checks to take the place of the glazed pots described above we employ the same pots coated with asphaltum paint. These are impervious to diffusible salts as shown by our tests in the laboratory. It is hoped that the method outlined here will be tested by other investigators in the study of important soil problems.

SOME BACTERIOLOGICAL RELATIONS IN SOILS KEPT UNDER GREEN-HOUSE CONDITIONS.

By JACOB G. LIPMAN and IRVING L. OWEN.

Soils in the green-house are exposed to conditions that are admittedly more or less artificial. The range of temperature and moisture is not the same as that in field soils, while the aeration of green-house soils is, if anything, even more artificial. The artificial conditions are emphasized still more strongly when the soil is kept in small pots. The operations incident to the filling of the pots and the applications of fertilizers involve a more intimate contact of the soil particles with atmospheric oxygen than is possible under field or garden conditions. This leads to an abnormal multiplication of the soil bacteria and to a consequent abnormally rapid oxidation of the organic matter. In the course of time the more readily decomposable portions of the organic matter in the soil become depleted and this is followed, in turn, by a decline in the numbers of bacteria that will grow on agar plates. It is possible that the rapid falling off in numbers is due not merely to the depletion of the readily decomposable organic matter, but also to the accumulation of certain cleavage products injurious to the bacteria. Of the latter factor we have no direct knowledge and it is referred to in this place only as a possible explanation of the facts recorded below. It may be added here, also, that the greatly decreased number of bacteria appearing on agar plates should not be accepted as absolute proof that the total number of microorganisms in the soil had diminished. There is a possibility that a compensating increase had occurred of bacteria that do not grow on agar plates, as for instance, the nitrous and nitric ferments, etc.....

With the foregoing observations as an introduction we may now turn to the examination of some data concerning quantitative bacteriological relations in soils kept under green-house conditions. The

experiments under consideration were carried out either in earthenware or glass pots each containing 20 pounds of soil. The various soil portions received different chemical or bacterial treatment, since it was the purpose of the experiments to determine whether such treatment would affect the numbers of bacteria producing colonies on agar plates.

Series I.

The effect of varying quantities of acid phosphate and of citric acid on the number of colonies on agar plates.

Fourteen portions of fertile soil were placed in large tarred glass pots and maintained at a water content of 14 per cent. No crop was grown on these soils. The several soil portions were treated as follows:

Pot No.	Material applied	Colonies on Agar plates per gram of soil			
		Dec. 10	Dec. 21	Jan. 14	Feb. 4
1	18·14 gms. carbonate of lime	6,000,000	6,200,000	4,000,000	3,200,000
2	0	7,920,000	6,000,000	4,460,000	2,600,000
3	0·1496 gms. acid phosphate	6,000,000	7,800,000	4,240,000	2,640,000
4	0·2992 ,, ,,	4,600,000	7,300,000	3,900,000	2,280,000
5	0·5984 ,, ,,	3,640,000	4,720,000	3,400,000	2,180,000
6	1·1968 ,, ,,	7,600,000	7,400,000	4,140,000	2,640,000
7	1·7952 ,, ,,	4,480,000	5,340,000	4,400,000	2,840,000
8	18·14 gms. carbonate of lime	6,400,000	7,840,000	3,700,000	2,860,000
9	0	7,120,000	7,800,000	3,900,000	2,800,000
10	0·3233 gms. citric acid	6,840,000	6,400,000	4,600,000	3,000,000
11	0·6466 ,, ,,	9,400,000	5,500,000	4,040,000	3,300,000
12	1·2932 ,, ,,	14,400,000	13,000,000	4,700,000	3,920,000
13	2·5864 ,, ,,	11,400,000	9,230,000	5,100,000	3,320,000
14	3·8796 ,, ,,	12,600,000	10,300,000	7,200,000	3,680,000

The experiment was begun on November 17, and about three weeks later the first samples of soil were drawn for quantitative examination. The soil suspensions for plating were prepared by shaking 100 gms. of soil with 200 c.c. of sterile water and making the proper dilution for inoculation into synthetic agar[1]. The plates were kept in the incubator at 28° C. and counts were made at the end of three days. Plates were similarly prepared from samples drawn on December 21, January 1st and February 4th.

In comparing the results secured with the first set of samples we observe that the acid phosphate caused a gradual decrease in numbers when 0·1496 gms., 0·2992 gms., and 0·5984 gms. were used. When the amount of acid phosphate was further increased to 1·1968 gms. there was an increase, rather than a decrease, in the number of colonies. A

[1] *N. J. Sta. Rep.* 1908, p. 133.

still further increase in the amount of acid phosphate used was again followed by a decrease. It seems, also, that the application of lime depressed the number, since pots 2 and 9 to which nothing was applied, contained 7,920,000 and 7,120,000 bacteria, respectively, per gram of soil; while pots 1 and 8 that had received additions of carbonate of lime contained only 6,000,000 and 6,400,000 bacteria, respectively, per gram of soil. We observe, likewise, that the addition of citric acid stimulated the multiplication of the bacteria three weeks after the beginning of the experiment. With 0·3233 gms., 0·6466 gms. and 1·2932 gms. of citric acid the numbers of bacteria per gram of soil were 6,840,000, 9,400,000, and 14,400,000 respectively. Beyond that the increased application of citric acid led to an increase and then to a decrease in the number of bacteria.

On examining the results secured with the samples of December 21, we note corresponding relations in most instances. The applications of citric acid still showed their influence in the increased number of bacteria in most of the soils; while the acid phosphate showed an increase in some cases and a decrease in others. On the whole, however, we note a tendency towards smaller numbers in the soils that had received additions of citric acid; and a tendency towards greater numbers in the soils that had received additions of acid phosphate.

In the samples of January 14th we already observe not only smaller numbers of bacteria in all cases, but a distinct tendency towards the obliteration of the differences noted in the samples of December 10th, and December 21st. Nevertheless, we can still detect even here the depressing effect of some of the smaller applications of acid phosphate; and the stimulating effect on bacterial growth, of the applications of citric acid. Thus the minimum number of bacteria was found in soil 4 that had received an application of 0·5984 gms. of acid phosphate; and the maximum number in soil 14 that had received an application of 3·8796 gms. of citric acid.

Finally, in the samples of February 4th we find a still more marked decline in numbers and pronounced tendency towards the establishments of uniformity in so far as the number of colonies on agar plates was concerned. The influence of the applications of acid phosphate is scarcely apparent here, while the influence of the applications of citric acid is shown to a comparatively slight extent. There is no doubt, therefore, that in the interval between December 21st and February 4th the numbers of bacteria in all of the soils under experi-

ment decreased very rapidly either on account of the depletion of the available organic nutrients, or on account of the accumulation of injurious cleavage products.

Series II.

Influence of small additions of fertile soil on the bacterial content of quartz sand properly supplied with plant-food.

The present series consisted of eight tarred glass pots each containing 19·8 pounds quartz sand and 0·2 pounds of fertile soil. In some instances the fertile soil was sterilised previous to the mixing with the sand. Four of the soils were to be seeded to oats, the other four, exactly alike in treatment, were to remain uncropped. The fertilizer additions to each soil consisted of:

4·0 gms. acid phosphate
2·0 ,, muriate of potash
0·5 ,, magnesium sulphate
0·01 ,, ferric sulphate
2·0 ,, dried blood
5·0 ,, ground oyster-shell lime

The special treatment of the soils was as follows:

Pot No.	
1	Unsterilised soil, oats
2	,, ,, ,,
3	,, ,, uncropped
4	,, ,, ,,
5	Sterilised soil, oats
6	,, ,, ,,
7	,, ,, uncropped
8	,, ,, ,,

The oats were seeded on December 2 and harvested on April 2nd. They were found to contain the following amounts of dry matter and of nitrogen:

Pot. No.	Dry matter, gms.	Nitrogen, %	Nitrogen, mgs.	Average, mgs.
1	6·5	1·19	7·73	
2	5·2	1·42	7·38	7·55
5	9·2	1·51	13·89	
6	5·0	1·52	7·60	10·74

In pot 5 that had received an addition of sterilised soil the yield of dry matter and of nitrogen was considerably larger than that in the corresponding pot 6. It will be noted, also, that the sterilisation of the soil led to a larger proportion of nitrogen in the dry matter. This is not at all strange, for the sterilisation of the soil in the autoclave causes the unlocking of some of the inert nitrogen compounds.

Bacterial counts were made in samples drawn December 15th, January 4th and February 25th. Synthetic agar was used for plating and the colonies were counted at the end of two days. Check counts made at the end of four days revealed larger numbers, but the results given here are those obtained at the end of two days. They are strictly relative and agree with the check counts. The numbers of bacteria per gram of soil as found at the different dates were as follows:

Pot. No.	December 15	January 4	February 25
3	7,000,000	3,700,000	840,000
4	9,400,000	3,480,000	780,000
7	3,780,000	7,000,000	1,200,000
8	4,420,000	8,500,000	1,178,000

It is apparent from the counts recorded above, that the unsterilised soil caused a more rapid growth of the microorganisms at the beginning. About two weeks after the initiation of the experiment pots 3 and 4 contained, on the average, more than eight millions of bacteria per gram of soil, while the corresponding pots 7 and 8 contained slightly more than four millions in the same quantity of soil. But in the samples of January 4 we find these relations reversed. The soils of pots 3 and 4 contained, then, about 3·5 millions of bacteria per gram, while those of pots 7 and 8 contained 7·75 millions per gram. In the samples of February 25th the numbers had declined to a very marked extent, but the soils of pots 7 and 8 still contained a larger number of organisms than those of pots 3 and 4. We find, thus, that the sterilisation of the soil, by eliminating the bacteria, furnished less favourable bacteriological conditions for the decomposition of the organic matter. However, this held good only for a short time. After that there was a more intense development of the microorganisms, a more rapid decomposition of the organic matter favoured by its exposure to a higher temperature and pressure in the autoclave, and a more abundant supply of available nitrogen compounds to the crop.

Later still there was a falling off in numbers both in the unsterilised and sterilised soil. In the samples of February 25th, the numbers were reduced to less than one million per gram of soil in pots 3 and 4, and to slightly more than one million in pots 7 and 8. Evidently the mineralisation of the organic matter was proceeding rapidly and the numbers were decreasing with the decreasing supply of available food. Hence, the results of the present series agree completely with those of the preceding series.

Series III.

The numbers of bacteria in green-house soils as affected by additions of organic matter and of cultures of B. Mycoides.

Like the preceding, this series consisted of eight tarred glass pots. Four of them were filled with 20 pounds of quartz sand, and the other four with 20 pounds of fertile soil. The quartz sand received in each case additions of:

4·0 gms. acid phosphate
2·0 ,, muriate of potash
0·5 ,, magnesium sulphate
0·01 ,, ferric sulphate
2·0 ,, dried blood
5·0 ,, ground oyster-shell lime

The fertile soil received additions of 5·0 gms. of oyster-shell lime only. Aside from the materials enumerated some of the pots received additions of 0·2 pounds of green grass, and unsterilised or sterilised cultures of *B. mycoides*. Twenty-five cubic centimetres of a three days old culture of the latter was employed in each case. The various additions are shown in the following table:

	Pot No.		
Quartz sand	1	Fertilizer, lime	Sterile culture, *B. mycoides*
	2	,, ,,	Live ,, ,,
	3	,, ,, green grass	Sterile ,, ,,
	4	,, ,, ,,	Live ,, ,,
Fertile soil	5	Lime	Sterile ,, ,,
	6	,,	Live ,, ,,
	7	,, green grass	Sterile ,, ,,
	8	,, ,,	Live ,, ,,

The experiment was begun on December 8th and samples for plating were drawn on January 7th, February 2nd, March 5th and March 29th. Synthetic agar was employed as in the preceding experiments. The plates were kept in the incubator at 28° C. and their colonies counted at the end of three days. The numbers found per gram of soil were as follows:

	Dates of sampling			
Pot No.	Jan. 7	Feb. 2	March 5	March 29
1	4,000,000	240,000	1,200,000	34,000
2	4,480,000	280,000	1,100,000	56,000
3	6,400,000	320,000	1,000,000	30,000
4	4,960,000	260,000	1,060,000	52,000
5	2,000,000	4,520,000	4,400,000	800,000
6	2,440,000	2,000,000	4,600,000	980,000
7	6,120,000	1,640,000	5,200,000	1,280,000
8	4,000,000	1,480,000	6,000,000	840,000

The samples of January 7th show the effect of the grass in both the quartz sand and in the fertile soil. In the former, the average number of bacteria per gram was 4,240,000 where no grass was used, and 5,680,000 where grass was used. The corresponding numbers in the fertile soil were 2,220,000 and 5,060,000. Aside from the influence of the grass we note that the additions of live cultures of *B. mycoides* increased the numbers in both the quartz sand and the fertile soil when no grass was employed, and decreased the numbers when grass was used. It is also interesting to note that at this time the quartz sand contained a greater number of bacteria than were present in the fertile soil.

On February 2nd the quartz sand had become very poor in bacteria. The average number at that time was less than 300,000 per gram, and the influence of the additions of grass was scarcely apparent. In the fertile soil, on the other hand, the numbers of bacteria were maintained at a higher level. In fact, in pots 5 and 6 where no additions of grass were made the average number of bacteria was greater on February 2nd than it was on January 7th. In pots 7 and 8, on the contrary, there was a very marked falling off in numbers from the earlier to the later date. We may observe, likewise, that the soils of pots 3, 5, and 7 which had received additions of sterile cultures of *B. mycoides* contained larger numbers of bacteria than the corresponding soils 4, 6, and 8 that had received cultures of live bacteria.

When we compare the numbers found in the different soils on March 5 we observe, in the first place, an appreciable increase in all but one case. The increase is particularly marked in the quartz sand where the numbers rose from less than 300,000 to more than 1,000,000 per gram. An even larger increase occurred also in soils 7 and 8 where the corresponding numbers rose from a little over 1·5 millions to more than 5·5 millions. It seems, also, that the soils which had received additions of live cultures of *B. mycoides* showed a tendency towards greater numbers than those present in the corresponding soils that had received additions of sterilised cultures. This is contrary to the results secured with the samples of February 2.

The samples drawn on March 29th contained much smaller numbers of bacteria than those present in the other samples. In the quartz sand the numbers were reduced, on the average, to less than 50,000 per gram; while in the soil they were reduced to less than 1,000,000 per gram. The influence of the additions of grass was no longer marked at this date. It appears, therefore, that in the quartz

sand the additions of live cultures of *B. mycoides* (pots 2 and 4) was not without influence on the numbers found on March 29th. Generally speaking, therefore, we observe in this as in the preceding series an unmistakable decline in the numbers of bacteria in the soil, or at least of those bacteria that form colonies on agar plates under aerobic conditions.

Series IV.

The influence of gypsum on the numbers of soil bacteria that form colonies on agar plates.

Twelve portions of fertile soil each weighing 18 pounds were placed in earthenware pots and treated as follows:

Pot No.	Treatment	Yield of dry matter	Average
1	0	16·2 gms.	
2	0	6·2 ,,	11·2 gms.
3	2 gms. gypsum	17·7 ,,	
4	2 ,,	5·2 ,,	11·4 ,,
5	4 ,,	10·8 ,,	
6	4 ,,	6·3 ,,	8·5 ,,
7	8 ,,	20·2 ,,	
8	8 ,,	10·7 ,,	15·4 ,,
9	0	—	—
10	2 gms. gypsum	—	—
11	4 ,,	—	—
12	8 ,,	—	—

The soil in pots 1, 3, 5, and 7 was seeded with vetch; that in pots 2, 4, 6, and 8 with oats. The soils in pots 9—12 remained uncropped and were sampled from time to time for bacteriological examination.

In looking over the yields of dry matter as given in the foregoing table we find that the 2 gms. of gypsum produced no appreciable increase or decrease. The 4 gms. of gypsum apparently reduced the yields, but on closer examination we find that the decrease occurred in the vetch and not in the oats. On the other hand, the 8 gms. of gypsum produced an unmistakable increase in both the vetch and the oats. There is reason to think, therefore, that the applications of gypsum were not injurious to the soil bacteria, nor to the plants themselves. The larger yields of dry matter in the soils that had received additions of 8 gms. of gypsum may be partly, if not wholly, accounted for by the larger amounts of nitrate furnished to the plants in these soils. This is evidenced by the nitrate determinations made on March 1st and April 26th. The following amounts were found:

Pot No.	Dates of sampling: March 1	April 26
9	0·612 p.p.m.	5·200 p.p.m.
10	1·780 ,,	10·410 ,,
11	3·450 ,,	6·250 ,,
12	2·880 ,,	15·620 ,,

We observe from the foregoing that the amounts of nitrate nitrogen expressed in parts per million, were favourably effected by the additions of gypsum. The untreated soil contained the smallest amount of nitrate nitrogen both on March 1st and April 26th. In the treated soils the increase due to the gypsum was not always regular, though unmistakable. For instance, less nitrate was contained in soil 12 than in soil 11 on March 1; while on April 26 the nitrate content of soil 11 was smaller than that of soil 10. Whatever the cause of this irregularity it is clear that the gypsum promoted nitrification and furnished thereby a better supply of available nitrogen to the plants.

As to the effect of the gypsum on the bacteria forming colonies on agar plates we find the following:

Pot No.	Dates of sampling: Feb. 25	March 29	April 26
9	2,520,000	300,000	1,260,000
10	2,800,000	820,000	980,000
11	3,360,000	560,000	960,000
12	3,200,000	1,000,000	960,000

The samples of February 25 show that the gypsum increased the numbers of the bacteria growing on agar plates. The maximum increase occurred in soil 11 where there were present 3,360,000 bacteria per gram as against 2,520,000 in soil 9. A month later the numbers of bacteria in these soils were much smaller. In soil 9 the number was reduced to 300,000 per gram, and in soil 11 to 560,000 per gram. For all that, the influence of the gypsum was still felt. But when we compare the numbers found on April 26th, we note an increase rather than a further decrease, in three out of the four soils. Nevertheless, the results before us confirm those secured in other series. There is in each case a pronounced tendency towards a falling off in the numbers of bacteria that will form colonies on agar plates.

Considered in their entirety the results of these experiments prove clearly that in green-house soils there may be at first a very rapid increase of decay bacteria to numbers above the normal and then a gradual decline to numbers decidedly below the normal. It seems, also, that the nitrifying organisms become more prominent as the others gradually decrease in numbers. Furthermore, there are indications of periodicity in the increase and decrease of the decay bacteria in the soil; and the possibility is not excluded that with a much longer

period of observation than that allotted to the present experiments the numbers of bacteria producing colonies on agar plates would have risen again to very considerable proportions.

As to the cause of the decrease in numbers it may lie, as already suggested, in the gradual mineralisation of the organic matter and the partial exhaustion of the more readily available organic food. It is less probable that the decrease may be due entirely to the accumulation of injurious cleavage products, although this accumulation may serve as a contributing cause. It is conceivable how the readily decomposable organic food may be laid fast in the bodies of the microorganisms, and how, on the death and decomposition of the latter the organic matter may again become available for the growth of other bacteria. This assumption would account for periodicity in the increase and decrease of decay bacteria in the soil. Moreover, the partial mineralisation of the organic matter and the accumulation of nitrate would favour the growth of algae which, in turn, would provide an additional amount of organic nutrients and encourage, thereby, the multiplication of the decay bacteria. The algae would thus become one of the factors responsible for periodicity in the increase and decrease not only of decay bacteria, but also of nitrifying and of nitrogen fixing bacteria.

Generally speaking, the abnormal multiplication and subsequent gradual decline in the numbers of soil bacteria may be brought about by a variety of factors. We know that intensive tillage will do it in soils not abundantly supplied with fresh organic material. We know, likewise, that the sterilisation of soil by heat or the more or less thorough drying out of the soil will be followed by a more intense multiplication of the decay bacteria. Similar effects may be produced by carbon bisulphide, chloroform, hydrogen peroxide and other volatile antiseptics or germicides. In some instances the abnormally large numbers may be due to the destruction of the species equilibrium, in others to the supply of an uncommonly large amount of available food; while in still other instances both of these may be prominent factors. But when all is said, we must conclude that it is the quantity as well as the quality of the available food that is of controlling moment in the growth of soil bacteria as it is in the growth of higher plants. We have still much to learn concerning the conditions under which the utilisation of available plant-food by bacteria may hinder the growth of crops. The competition for food between the higher and lower organisms may not always be favourable to the former, and it is therefore desirable to widen our knowledge so as to permit us by proper methods of soil treatment to turn the balance in favour of the cultivated plants.

ON THE QUANTITY OF AMMONIA AND NITRIC ACID IN THE RAIN-WATER COLLECTED AT FLAHULT IN SWEDEN.

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Numerous investigations have been made on the amounts of combined nitrogen in rain and snow in different parts of the world during the last 60 years.

The most extensive are those at Rothamsted, where the rain-water collected in 1853–6 and since 1878 has been analysed continuously up to the present time. The results are to be found in several papers published by the late Sir J. B. Lawes, Sir J. H. Gilbert and Mr R. Warington, whilst a recent paper by Dr N. H. J. Miller[1] contains a *résumé* of all the analyses up to the end of the harvest year 1904—1905, besides a complete bibliography of the papers published on this question in different countries since 1761.

The average amount of nitrogen in the forms of ammonia and nitric acid in the rain falling at Rothamsted during 13 harvest years 1888–9—1900–1, is 3·84 lbs. per acre per annum. The amounts found at 35 other stations, of which 20 are in Europe, during the last 40 years are also given in Dr Miller's paper.

Such investigations have not hitherto been made in Sweden, and we therefore began last year (1909) to collect the rain and snow at our experimental station at Flahult, and to estimate the amount of ammonia and nitrates in monthly samples[2].

[1] "The amounts of nitrogen as ammonia and as nitric acid and of chlorine in the rain-water collected at Rothamsted," *Journal of Agricultural Science*, 1906, **1**, 280.

[2] A more detailed paper on the subject has just been published in a Swedish journal. Nitrites are included with nitrates in our determinations.

The Station is situated in latitude 57°42′, is 14°8′ east of Greenwich, and 222·8 metres (730 feet) above the sea-level. The distance from the nearest town, Jönköping[1], and the Wettern Lake is 11 kilometres (7 miles), from the Baltic 150—160 kilometres (about 95—100 miles), and from the North Sea 130—140 kilometres (about 80—85 miles).

The prevailing winds are from the south-west and south; the average amount of rain during the last 7 years (1902–8) was 577 millimetres (22·72 inches), the number of days with rain or snow 162, and with snow alone 49. The rain gauge is 1·5 metres (5 feet) above the ground, 20 metres from the nearest building, and 40 metres east of the pine forest that surrounds the experimental farm.

The amount of rain or melted snow collected daily is put into a large glass bottle holding 20 litres, which at the end of the month is brought to our chemical laboratory in Jönköping and the ammonia and nitrates determined by the methods described by R. Warington[2].

In Table I. are given the analytical results, while Table II. contains certain meteorological data.

TABLE I.

1909	Rainfall	Nitrogen						
		Per million		Per acre			% of total	
		as NH_3	as N_2O_5	as NH_3	as N_2O_5	Total	as NH_3	as N_2O_5
	inches			lbs.	lbs.	lbs.		
January	1·63	0·371	0·266	0·137	0·098	0·235	58·2	41·8
February	0·57	0·593	0·148	0·076	0·019	0·095	80·0	20·0
March	2·43	0·741	0·151	0·407	0·083	0·490	83·1	16·9
April	1·56	0·403	0·167	0·142	0·059	0·201	70·7	29·3
May	2·32	0·556	0·231	0·292	0·121	0·413	70·6	29·4
June	4·01	0·494	0·128	0·448	0·116	0·564	79·4	20·6
July	5·24	0·267	0·128	0·317	0·152	0·469	67·6	32·4
August	3·25	0·618	0·185	0·454	0·136	0·590	77·0	23·0
September	3·28	0·502	0·180	0·373	0·134	0·507	73·6	26·4
October	3·42	0·377	0·180	0·292	0·139	0·431	67·7	32·3
November	1·52	0·466	0·206	0·160	0·071	0·231	69·3	30·7
December	3·32	0·283	0·229	0·213	0·172	0·385	55·3	44·7
Whole year	32·55	0·450	0·177	3·317	1·302	4·619	71·8	28·2

[1] Jönköping has 24,000 inhabitants. As the town is on the north side of Flahult and the winds are generally from the south and south-west, contamination of the rain with smoke, &c., can only be slight.

[2] "The amount of nitric acid in the rain-water at Rothamsted, with notes on the analysis of rain-water," *Journal of the Chemical Society*, 1889, **55**, 537.

TABLE II.

1909	Number of days with			Snow lying on the ground		Temp. of the air, average of month		Number of cloudy days	Days with thunder
	Rain	Snow	Total	Number of days	Average depth	1909	1859—1900		
					inches	deg. F.	deg. F.		
January	3	13	16	15	4·72	26·7	25·7	21	—
February	1	7	8	28	6·30	21·2	25·2	8	—
March	4	14	18	31	9·06	25·9	28·1	25	—
April	7	8	15	6	2·36	34·1	36·7	13	—
May	10	6	16	—	—	41·2	43·6	10	—
June	17	—	17	—	—	53·6	54·6	11	5
July	16	—	16	—	—	55·7	58·4	9	3
August	16	—	16	—	—	55·8	56·1	6	1
September	15	—	15	—	—	48·0	49·1	12	1
October	24	—	24	—	—	46·9	40·6	17	—
November	9	10	19	17	1·97	28·5	33·3	12	—
December	7	10	17	15	6·30	28·1	27·5	25	1
Whole year	129	68	197	112		38·6	40·1	169	11
Corresponding numbers:									
Year 1908	118	48	166	85	—	39·8	—	159	12
„ 1907	133	55	188	81	—	39·6	—	167	4

The year 1909 showed several unfavourable meteorological conditions. The amount of rain exceeded the average of the last 7 years by more than 40 per cent., and the number of days with precipitation is also higher (197 against an average of 162 days).

The mean temperature of the air for the whole year was below the average of 42 years, and was only above the average in three months. In May and July especially low temperatures were recorded.

Turning to Table I. the amount of nitrogen in the rain and snow is seen to vary considerably from month to month. The nitrogen as ammonia varies between 0·267 parts per million in July to 0·741 in March, whilst the nitrogen as nitrates and nitrites varies from 0·128 to 0·266.

The relative proportions of ammoniacal and nitric nitrogen vary, but the averages for the whole year are 71·8 and 28·2 per cent. respectively of the total, corresponding very closely with the Rothamsted results.

The total amount of nitrogen in the rain and snow at Flahult during the year 1909 was 4·619 lbs. per acre. This again agrees very well with the result obtained at Rothamsted, and at some other places.

The analyses will be continued during the present year 1910.

AMOUNT OF COPPER IN TEA SPRAYED WITH BORDEAUX MIXTURE.

By H. E. ANNETT, B.Sc., F.C.S., *Officiating Agricultural Chemist to the United Provinces*, AND SUBODH CHUNDRA KAR, M.A., *Assistant to the Imperial Agricultural Chemist.*

As a result of a bad attack in tea gardens of blister blight caused by the fungus *Exobasidium vexans*, Mr McRae, the officiating Imperial Mycologist, was deputed by Government to combat the disease. Mr McRae has tried the effect of spraying the tea bushes with Bordeaux mixture on an experimental scale. In view, however, of the recent scare in America over the sale of grapes sprayed with Bordeaux mixture, it was thought desirable to have these spraying experiments under chemical control. Accordingly an area in a tea garden was set apart and the bushes sprayed with Bordeaux mixture. After nine days the tea was picked both from the sprayed and from an unsprayed area, and about 80 lbs. (a chest) of tea was manufactured from the produce of each area. Great care was taken to prevent contamination. Samples of tea both from the sprayed and unsprayed areas were carefully sealed up and sent to this office for analysis. None of this tea was put on the market.

On analysis the unsprayed tea was found to contain $\frac{1}{12}$ grain per lb., whilst the sprayed tea contained $\frac{1}{2}$ grain per lb. The analyses were duplicated, special precautions being taken to avoid any chance of copper contamination.

Methods used. 50 grms. of the sample were ignited in a muffle furnace and the ash extracted with nitric acid. The solution was evaporated to dryness and heated at 100° to render silica insoluble. The residue was taken up with water and a drop or two of hydrochloric acid and filtered. In this solution the copper was precipitated with sulphuretted hydrogen gas, filtered and the precipitate ignited and then dissolved in HNO_3. This solution was evaporated to dryness and taken

up with water and a few drops of diluted hydrochloric acid and filtered. From this solution the copper was again precipitated with sulphide of hydrogen, redissolved in nitric acid, and after evaporation again taken up with water and hydrochloric acid as before. This process was again repeated. In this solution the copper was determined colorimetrically first with ammonia and then the blue solution was evaporated to dryness and gently heated and then taken up with water and a little hydrochloric acid and the copper estimated colorimetrically with potassium ferrocyanide.

The two methods were found to give very concordant results and all the results obtained are here tabulated.

Description of tea	Weight of tea taken	Weight of copper found	
		Colorimetric ammonia method	Colorimetric K_4FeCn_6 method
Unsprayed	(1) 50 grms.	·0006	—
,,	(2) 50 grms.	·0006	·0007
Sprayed	(1) 50 grms.	·0034	—
,,	(2) 50 grms.	·0034	·0037

For the analyses set out in the following table, known amounts of copper sulphate were added to separate lots of 50 grms. of tea and the copper estimated by the two methods outlined above. As will be seen the amounts of copper found are in close agreement with the amounts added, thus showing the methods of analysis used to be trustworthy.

Weight of tea taken	Amount of copper added	Amount of copper found	
		Colorimetric ammonia method	Colorimetric K_4FeCn_6 method
50 grms.	nil	·0009	·0008
,,	nil	·0010	·0009
,,	·001 grms.	·0021	—
		·0019	·0020
,,	·002 grms.	·0028	—
		·0030	·0028

The tea used in these last experiments was of a brand well known on the market.

It is well known that copper in small amounts is commonly present in various food stuffs. Blyth[1] gives the following table showing the amounts of copper found normally in various foods:

	Mgrms. per kilo	Grains per lb.
Wheat	5·2—10·8	·037—·078
Rye	5	·037
Oats	8·5	·060
Barley	11·8	·086
Rice	1·6	·011
Bread	1·5—4·4	·011—·033
Vermicelli	2—10	·015—·075
Grouts	1·6—3	·011—·022
Potatoes	1·8	·012
Beans	2—11	·015—·082

Blyth goes on to say that in similar small quantities it has also been found in carrots, chicory, spinach, blackberries, peaches, pears, figs, plums, tamarinds, black pepper, and many other fruits and spices. The most common food which has a high copper content is cocoa which contains from 12—29 mgrms. per kilo (= ·09—·22 grains per lb.).

By our analyses it has been established that tea also contains an appreciable amount of copper.

Dr Leather very kindly carried out the following experiments to see how much of the copper present in the tea might actually be consumed by the tea drinker: 36 grms. of tea, which was found to be about equal to eight teaspoonfuls, were treated with two litres of boiling water for five minutes as in the ordinary process of making tea (eight breakfast cups hold about two litres water). Two experiments were carried out, one with unsprayed tea and one with the sprayed tea used in my experiments. The tea infusion was filtered off, evaporated to dryness and ignited. The ash was extracted with nitric acid, ammonia added in excess and the solution filtered from precipitated phosphates. The filtrate on examination in a Nessler tube showed no trace of blue colour in the case of the unsprayed tea. Sprayed tea gave a slight blue colour = 0·0002 gr. Cu, so that if one drank the whole eight cups of tea one would take into one's system an almost inappreciable amount of copper.

[1] A. W. Blyth, *Poisons and their detection.*

NOTE ON THE COMPOSITION OF GAS LIME.

By P. J. BHATT, B.Sc. (Bom.), L.Ag. (Bom.).

Very few analyses of gas lime have been published; consequently the following analyses made in the laboratory of the Department of Agriculture of the University of Cambridge may be of interest to readers of this *Journal*.

The samples of gas lime were obtained from the Cambridge Gas Works. One was what is known as carbonated lime, that is to say, lime which had been used for purifying the gas after it had previously been purified by hydrated ferric oxide. This sample, as shown by the analysis, is practically moist calcium carbonate. It was obtained in a perfectly fresh condition. The second sample was described by the Works Manager as "sulphided lime." It had been used for purifying gas without the previous use of ferric hydrate. A sample was taken from the centre of a large heap which had been standing for about a year.

The following method of analysis was used:

Moisture and loss on ignition were determined in finely ground samples in the ordinary way; bases were determined by evaporating a sample with hydrochloric acid to complete dryness, moistening the residue with hydrochloric acid, boiling with water, and filtering off the silica. Ferric oxide, aluminia, and traces of phosphates were precipitated from the filtrate by ammonia, and weighed together. Lime was precipitated with ammonium oxalate. The filtrate from this precipitate was evaporated to dryness, and the residue ignited. The magnesia was weighed as oxide, and the alkalies as chlorides. Carbon dioxide was determined by weight after absorption in caustic potash, the sulphuretted hydrogen having been previously absorbed in hot silver nitrate solution. Free sulphur was determined by extraction with carbon bi-sulphide, sulphocyanide by titration with standard potassium permanganate. Cyanides were estimated by precipitating

them from the water extract as Prussian blue, which was filtered off, dissolved in soda, and titrated with potassium permanganate.

The analyses are given below:

	Carbonated lime	Sulphided lime
Moisture	26·62	29·06
Silica	3·81	3·82
Fe_2O_3, Al_2O_3, etc.	1·86	1·99
CaO*	35·37	34·08
MgO	0·39	0·28
Alkalies as chlorides†	0·86	1·26
CO_2	24·74	23·65
H_2S	None	0·17
SO_3	Slight trace	2·66
Free sulphur	None	0·407
HCNS	Slight trace	0·710
HCN	None	0·18
Ether extract	None	0·488
* Free lime	1·825	None
† KCl	0·359	0·296
Loss on ignition	30·19	25·82

The analyses show clearly that gas lime possesses little fertilizing power beyond that of chalk. Carbonated lime seems to be quite innocuous. Sulphided lime contains small but definite quantities of free sulphur, sulphur compounds, and cyanides, which may be harmful to crops or possibly may give the substance some fungicidal power.

To test these two points, solutions were made from each sample by shaking in 100 c.c. of water 0·1, 1·0, 10·0 grams. These solutions were then used to moisten cress and clover seeds which were germinating in Petri dishes on filter paper. The rate of germination and the growth of the seedlings was observed for some days. The average length of seedlings grown in the various solutions was found to be:

	Average length of seedlings grown in						
	Pure water	Carbonated lime			Sulphided lime		
		0·1 %	1·0 %	10 %	0·1 %	1·0 %	10 %
Cress	19	21	17	10	19	17	9
Clover	32	31	28	19	29	32	12

It will be seen that in the weaker solutions the seedlings grew practically as well as in pure water. The strongest solution however

was distinctly a check on growth, especially in the case of the sulphided lime.

A further trial was made by spraying solutions of the strengths mentioned above on to the leaves of seedlings of cress growing thickly in pots.

The stronger solutions produced some slight amount of scorching, but did no permanent damage.

It may, therefore, be concluded that neither of the samples of gas lime examined were likely to prove dangerously poisonous to crops growing on land to which they might be applied.

The next point was to test the possible fungicidal power of the samples. This was done by dissolving in the solutions of the strengths described above, the requisite amounts of sugar and salts for the growth of fungi. Hanging drops of each solution were then inoculated with the spores of *Penicillium glaucum*, and examined from time to time.

Duplicate experiments were also made with larger volumes of the solutions placed in watch glasses and kept in a moist chamber.

In both cases it was found that the solutions of carbonated lime had no appreciable effect on the germination and rate of growth of the spores.

A 1·0% solution of the sulphided lime was found to retard germination and growth very considerably, and a 10% solution prevented most of the spores from germinating.

Apparently, therefore, sulphided gas lime has a certain fungicidal power, due in all probability to the sulphur compounds and cyanides it contains, and possibly to the small amount of tarry ether extract.

In the sample examined, however, the fungicidal power does not seem to be strong enough to give the substance any practical value.

Finally, I must not omit to thank Professor T. B. Wood, at whose suggestion I took up this work, for his advice and guidance.

THE ACTION OF PLATINUM BLACK ON FREE NITROGEN.

By Dr OSKAR LŒW.

The attention of the writer was called recently to a publication of E. J. Russell and Norman Smith in this *Journal*[1], stating among other things that the observation of the writer of the production of nitrous acid and to a small amount of ammonium nitrite by platinum black in presence of alkaline bases as potash or baryta and air[2] could not be realised by those authors. Since this failure was considered by others as a refutation of my observation, a few lines of explanation may be in order.

In the first place it is necessary not to operate with too small quantities of platinum black; the writer applied generally 20—80 grams.

In the second place it is indispensable that the platinum black is of the utmost fineness of the particles and perfectly pure. Coarseness of particles, as resulting from certain methods of preparation, and further even small amounts of impurities, as *e.g.*, especially chlorides, render the platinum black unfit for catalytic processes of a more subtle nature[3].

Utmost care and precaution are especially required in carrying on the experiment above mentioned, since the nitrous acid reaction of *Griess* can be obtained almost with every object in a laboratory, where many gas flames producing it continuously are in use, as the writer has mentioned over twenty years ago. Also the caustic potash and soda bought as "chemically pure" sometimes contain traces of nitrite. Nobody who has read the communication of the writer will deny, that every precaution had been regarded.

[1] *Journ. Agric. Science*, I. p. 444 [1906].

[2] *Berichte der deutschen chemischen Gesellschaft*, vol. XXIII. p. 1443.

[3] A very deteriorating effect was, *e.g.*, also produced by treatment of the platinum black with chloroform.

As to the efficacy of platinum black, Doebereiner had already observed more than 60 years ago great differences, depending upon the fineness of the particles and the influence of minute amounts of impurities. A preparation of great efficacy is obtained by the method of the writer[1], which was recognised by others, as e.g. by A. Bringhentis in his treatise: "Catalysis and Electromotive Power."

By means of platinum black thus prepared, by formaldehyde and potash from platinum chloride, Prof. L. Woehler of Karlsruhe was also able to confirm my observation on the action of platinum black on atmospheric nitrogen[2].

The writer may add a further observation. Platinum black freshly prepared and well washed was kept slightly moistened in a well closed bell-jar in a room in which neither chemical work was carried on, nor gas was ever burnt, thus avoiding the danger of presence of nitrous acid in the air. After two months this platinum black yielded a strong blue reaction with diphenylamin-sulphuric acid, but not the red reaction of Griess, thus showing that nitric acid but not nitrous acid was formed under that condition. Nessler's reagent disclosed only very faint traces of ammonia[3]. Thus we succeeded in a merely catalytic way at the ordinary temperature to imitate the work of certain bacteria, as Azotobacter, in the soil.

[1] *Berichte der deutschen chemischen Gesellschaft*, vol. XXIII. p. 289.

[2] *Ibid.* vol. XXXVI. p. 3479.

[3] O. Lœw and K. Aso, *Bull. College of Agriculture*, vol. VII., University of Tokyo.

THE SYMPTOMS OF INTERNAL DISEASE AND SPRAIN (STREAK-DISEASE) IN POTATO.

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Introduction.

Frank[1] in 1897, and again[2] in 1898, describes an obscure potato disease under the name of "Buntwerden" or "Eisenfleckigkeit." The names, which mean rust or iron spots respectively, refer to the markings in the flesh of the tuber. This disease shows no characteristic external symptoms and can only be detected by cutting the tuber. The markings take the form of specks or lines which are not connected into a system and do not communicate with the surface as in the case of the brown markings due to the presence of Phytopthora. Frank was unable to detect the presence of a hyphal organism in the diseased tissue. He states that the disease does not spread during the storage period and that diseased tubers when planted give rise to perfectly healthy ones. Frank very clearly distinguishes between "Buntwerden" or "Eisenfleckigkeit" and "Trockenfäule" and considers that the former is due to prevailing conditions of soil and climate.

A troublesome disease, similar in many respects to "Buntwerden," has been prevalent for many years in this country and is attracting attention at the present time on account of the damage to crops for which it is actually or reported to be responsible. The symptoms are internal and on this account the disease is difficult to eliminate since it is impossible to select disease-free tubers at sight. Just as in

[1] Frank, *Kampfbuch gegen die Schädlinge unserer Feldfrüchte* (1897), p. 212.

[2] Frank, "Untersuchungen über die verschiedenen Erreger der Kartoffelfäule," *Ber. Deut. Bot. Gesell.*, Bd, xvi. (1898), p. 287.

"Buntwerden" or "Eisenfleckigkeit," the flesh is marked with brownish blotches or streaks and there are no traces of the presence of a hyphal organism in the tissue.

Very little seems to have been written on the subject in this country. One of the first references[1] is to be found in the "Report on Field Experiments conducted by the Agricultural Department of University College, Reading," issued in 1896. With regard to a sample of potatoes forwarded by Colonel Cornwallis West, Gilchrist and Foulkes state: "The potatoes were all badly affected with what is known as 'internal disease,' which Professor Marshall Ward, advisory expert in such subjects to the College, is now investigating. He finds that the tubers are badly affected with what the Germans call 'Trockenfäule,' which is believed (on insufficient evidence as yet) to be due to a bacterium." Analyses of soils, from different centres where considerable harm had been done to the potato crop by Internal Disease, are given, showing that the soils are exceptionally poor in phosphates, potash and lime; being moorish, sandy soils of a poor character resting on the Bagshot sands in the Ascot district.

Some years later, Messrs Sutton and Sons[2], in a Report dealing with potato experiments, state with regard to Internal Disease: "The proportion of diseased tubers in this year's crop does not appear to be in any way influenced by the presence or absence of disease in the tubers planted." This seems to me an exceedingly important observation.

A short note[3] entitled "Sprain in Potatoes" appeared in the *Journal of the Board of Agriculture* in April, 1909. The symptoms described are those characteristic of the Berkshire disease. The author of the article states that at least one case is reported of an affected field where potash and lime are not lacking. A further report[4] on "Sprain" appeared in the November number and it is there recognized that the symptoms are those of Internal Disease.

In the North of England and in Scotland there is a form of flesh-disease in which the rust-marks are streak-like. Tubers in this condition are often said to be "sprained" (figs. 1—4). The term "sprain" is used here in this limited sense, but in order to avoid confusion the name "Streak-disease" is introduced and may prove more convenient. It seems advisable to keep Internal Disease and Sprain (Streak-disease)

[1] Gilchrist and Foulkes, *Report on Field Experiments* (1895), pp. 19, 32.

[2] Sutton and Sons, *Potato Demonstration* (1906), p. 29.

[3] *Journ. of the Board of Agric.* Vol. XVI. (1909), p. 37.

[4] *Ibid.* p. 647.

distinct for a time since the recorded observations for the two forms do not yet strictly agree; moreover, blotch-like markings only and streak-like markings only are characteristic of large samples of potatoes of known variety as shown in the following table:

Date	Variety of Potato	Percentage affected	Character of the markings
1909	"Duchess of Cornwall" ...	11	Streaks
	"Edina"	16	,,
	"British Queen"	9	,,
	"The Sutton Flourball"...	33	Blotches
1910	"Duke of York"	20	Streaks
	"British Queen"	20	,,
	"Sharpe's Express".....	15	Blotches

Symptoms of Internal Disease.

The flesh of tubers affected with Internal Disease is marked with brown or chocolate-coloured blotches which may or may not form a connected system. Where the disease is strongly developed the blotches are numerous and vary in form and size (figs. 5, 6), but where only slightly developed these are often reduced to mere specks. The blotches are not confined to the stalk end of the tuber nor do they appear to spread from this end. They do not as a rule spread from the vascular bundles although the latter are often discoloured. The presence of the disease has been detected as early as July in very young tubers grown in Devonshire. When the tubers were examined it was noticed that the skin (cork) was discoloured in places, an appearance due to small areas of brownish tissue immediately beneath the cork cells. Even at this early stage the blotches were generally distributed in the potato.

The affected cells, which still retain their starch, are arranged in irregular groups. The protoplasm is of a granular appearance and brownish colour owing to the formation of a gummy substance within the cell. Sooner or later a wound-cambium is formed enclosing the discoloured and dead areas at the expense of the starch of the storage-cells surrounding the wound-cambium so that a zone of starchless cells surrounds the wound-cork. The walls of the wound-cork and the middle lamella of the diseased cells react strongly with Phloroglucin but the brown substance in the cell is only slightly coloured with this reagent. The change in the middle lamella indicated by the Phloroglucin reaction

appears to be a slow one which takes place after the death of the cell. At the same time no easily-observed disintegration of the cell-wall takes place in early stages, so that if the disease is propagated from cell to cell this does not involve decay of the wall at the time of the death of the cell.

A short time ago, the writer[1] announced the presence of an obscure organism in the cells of potatoes affected with Internal Disease and Sprain (Streak-disease) on account of the frequent appearance of spongy spheres and globular bodies in the diseased cells, in the cells of the wound-cork surrounding diseased areas and in the surrounding storage tissue. The bodies, by a process of budding, appear to give rise to progressively smaller bodies until excessively small units are reached. The appearances presented in the cells simulate plasmodia, sporangia, spores, zoospores, etc., to a remarkable degree. It has not yet been possible to show that the buds are definitely cut off from or multiply apart from the parent body, or that they grow when apart from the cell. The globular bodies often disappear in weak potash, but the spongy spheres have proved insoluble in strong acid and alkali. These often reach 40 micra in diameter—comparable in size to the "sporangia" of *Chrysophlyctis endobiotica*. The smallest bodies are sometimes less than one micron in diameter. For these reasons and moreover because bodies of a still more complicated character have been observed[2] in the Bracken (*Pteris*) there is some considerable doubt as to the real nature of the bodies in question. In "The Sutton Flourball" variety of potato, globular bodies (pink) and spongy spheres (yellow) are found restricted, in normal potatoes, to a small area of tissue at the base of the "eye." Since these bodies could be traced from the "eye" to the discoloured areas in diseased tubers it was thought at the time that the bodies, which normally existed endophytically, became under certain circumstances definitely aggressive. The discovery that very young tubers might be affected with Internal Disease afforded an excellent opportunity of testing for the presence of a parasite, but no organism of any kind was detected in the discoloured areas. The bodies in question, therefore, whatever their nature, appear to be an effect rather than a cause of the disease.

Frank's statement that the markings do not, as in the case of Phytopthora, reach the surface of the tuber possibly requires modifying.

[1] Horne, A. S., *Annales Mycologici* (1909), p. 286.

[2] Horne, A. S., "On the spongy bodies, spheres, and globular bodies present in the Bracken (*Pteris*) and Potato," *Centralblatt f. Bakteriologie*, Abt. II Bd. 27, 1910.

It should not be taken as satisfactorily established. The blotches very often occur immediately subjacent to the cork. In the immature tubers examined, diseased cells could be traced to within two or three cells of the cork-cambium. Very often in combined attacks of Phytopthora and bacteria the actual point of penetration is not easy to find. Once an entry is effected, the parasite is concerned more with the storage tissue than the cork and spreads in the former.

Symptoms of Sprain (Streak-disease).

The marks, in typically developed cases of Streak-disease, take an arc-like or wavy course in section and frequently form a series of curves, one within the other (figs. 1—4). The streaks sometimes form a connected system—in one case a potato was cut into slices 2 mm. thick, diagrams were made of all the slices and the outline of the diseased tissue reconstructed. Isolated cells and cell-groups occur as observed by Frank in "Buntwerden," but it is not always easy to be certain that cells or areas are not connected with others. In very slight attacks the markings may be reduced to short lines or specks. My experience at present is that samples of potatoes of known variety affected with Streak-disease may show transitional forms between the specked and streaked condition, but not between a specked or spotted and the blotched condition of tubers affected with Internal Disease. The course followed by the streaks in Streak-disease is at present quite unintelligible since the lines do not follow the path of the vascular bundles nor of any known arrangement or grouping of cells. It is equally difficult to understand why the markings should take the form of streaks in the one case and blotches in the other.

The cells forming the streak are connected but their arrangement is not very regular. No difference has been observed in the structure of the wall separating affected cells which would account for the course taken by the lines. The appearances presented by the diseased cells in Streak-disease and Internal Disease are very similar. Apparent cases of wall-alteration in early stages were generally due to fragments of the altered protoplasmic membrane of an adjoining cell. The middle lamella of the affected cells yielded either a slight or no reaction with Phloroglucin. After the development of wound-cork, the enclosed cells were often crushed owing probably to the pressure exerted during the activity of the wound-cambium. The middle lamella then reacted more strongly and the "gummy" matter within the cell feebly with the reagent.

The streaks in tubers badly affected with Streak-disease often extend to the edge of a cut slice and appear to end at the surface (figs. 1, 2, 4). The diseased cells generally reach to within a few cells of the cork. Less frequently they can be traced right up to the cork, to a small lenticel or to the ragged edge of a slight abrasion: the connection with the skin being maintained by a straggling line of cells which can be followed only by means of a series of sections.

Experiments were planned in order to test Frank's statement that tubers affected with "Buntwerden" when planted give rise to perfectly healthy tubers.

A. PLOTS IN DEVONSHIRE—1909.

Experiment 1. Potatoes affected with Streak-disease received from Scotland were planted in plots previously used for celery and never before for potatoes. The soil was of medium character upon a sub-soil of sandy clay (Permian) and was not specially treated with manure. All the tubers in the sample were used (these were not all actually diseased), two sets were made from each (thirty-six in all), and left overnight in lime. In July five plants were dead or the sets had not developed. Forty-six tubers were received on October 5 (not the total crop) and examined with the following result:

Internal brown specks	3
„ (very slight)	
Various diseases	3
Apparently sound	40
Total	46

Experiment 2. Potatoes affected with Streak-disease, grown in Durham, but of Scotch origin, were planted and treated as in experiment 1. At the time of planting 9 per cent. of the sample were diseased. One hundred and eighty-six tubers were received and examined with the following result:

Internal brown marks (slight)	14 = 7·5 %
Phytopthora infestans ...	5
Various diseases	5
Apparently sound	162
	186

Experiment 3. Potatoes from Berkshire affected with Internal Disease were planted as in the experiments described above. As many

as 33 per cent. (44/132) of the sample were diseased at the time of planting. Thirty-three sets were planted, of these nine either did not develop or the plant arising from the set died. I received 207 tubers of which a large number were very small. The result of an examination was as follows:

Internal Disease,	bad	10 = 5 °/₀
„	„ slight	21 = 10 °/₀
„	„ traces	16 = 7·7 °/₀
Apparently sound		160
	Total	207

B. Plots in Armstrong College Garden, 1909.

Experiment 4. Diseased tubers were selected from a sample affected with Streak-disease raised in the county of Durham and planted in plots previously used for potatoes in ordinary garden soil not specially manured. One hundred and two tubers were examined at the end of September with the following result:

Streak-disease, bad	22 = 22 °/₀
Apparently sound	80
Total	102

Experiment 5. Equal numbers of diseased and disease-free sets of "The Sutton Flourball" variety were planted as in the last experiment, but since very few plants produced tubers of any size, it was hardly possible to make a comparison between the two lots. Some of the potatoes obtained from diseased sets were diseased but it was noticed in digging up a diseased set that the small tubers attached to the plant originating from it were not diseased.

Experiment 6. Sixteen disease-free tubers from Kent were planted in soil mixed with fragments of potatoes affected with Internal Disease and an equal number in soil without such treatment. The potatoes grown from both lots when lifted in September were free from disease.

Cases of Streak-disease and Internal Disease occurred in the garden only among potatoes raised from seed affected with these diseases.

Complication with Phytopthora and minor diseases.

Internal Disease and Streak-disease are often complicated by the presence of *Phytopthora infestans* or diseases of minor importance.

A sack of potatoes from Bishop Auckland, typically affected with Streak-disease, contained at the same time tubers with the flesh marked by lines extending from the surface inwards which looked as if a very fine wire nail had been thrust in, allowed to rust and pulled out again, the hole being filled up with loose starch grains. One of these lines is shown in fig. 7. Confusion and misunderstanding may very easily arise if only a few tubers are sent for examination. For instance, some potatoes sent to the Armstrong College were correctly diagnosed as affected with "Black-leg" ("Schwarzbeinigkeit"), but after a series of visits to the farm from which the sample came it was found that only an infinitesimal portion of the total damage to crop was due to this disease. Internal Disease and Phytopthora are very frequently associated —sometimes intimately associated. A large quantity of "sprained" potatoes from the neighbourhood of Berwick was affected in this way. Special attention should be given to the illustration (Plate XIX). Fig. 6 shows one of the Berwick potatoes entirely affected with Internal Disease; fig. 8, another of the same lot affected with both Phytopthora and Internal Disease (a zone of tissue with the fungus present extends round the margin of the tuber). The potato in fig. 10 is one of a sample, raised in Devon, affected with Phytopthora or Phytopthora and bacteria—the markings in the flesh of this particular tuber resemble somewhat the streaks in Streak-disease. The tuber in fig. 6 shows no *external* symptoms of the presence of disease.

ANALYSIS OF SAMPLES.

Date	Variety of Potato	Locality	Form of disease
1909	"The Suttou Flourball"...	Berkshire	Internal Disease
,,	"British Queen"	Durham county	Streak-disease
,,	"Second Early" type	Scotland	Streak-disease
1910	,, ,, ,,	,,	Internal Disease complicated with Phytopthora
1909	?	Ireland	Streak-disease
1910	"Duke of York"	Seed offered for sale in Northumberland of Scotch origin	
1909	"Edina"		Streak-disease
,,	"Duchess of Cornwall" ...		
1910	"British Queen"		
,,	"Sharpe's Express".....	... ...	Internal Disease
1909	?	Dunbar	Brownish blotches and hollows in the flesh (see fig. 9)
,,	"Up-to-Date"	Durham county	Phytopthora complicated with bacteria
,,	"Dalhousie".....		
,,	?	Devon	Phytopthora probably complicated with bacteria

Damage to Crops.

It is difficult to estimate the extent of damage to crops caused by Internal Disease and Streak-disease as specific forms of disease throughout the country, and this cannot be properly done until the more important reported cases have been thoroughly investigated. Marshall Ward, who was at one time investigating Internal Disease, appears to have left no published records of his observations but Professor D. A. Gilchrist who is familiar with the Reading district tells me that it has existed for a number of years often in an acute form and causing considerable damage to crops. Professor E. S. Salmon writes "I know that a considerable loss occurs through 'sprain,' it is a common sight in the Borough (London) to see potato merchants slicing potatoes with a penknife and rejecting loads where 'sprain' is conspicuous." With regard to this use of the term, Professor Salmon writes "I apply the term 'sprain' to spots, lines or flecks of dry brown tissue in the flesh of the potato—often the marks are quite small, mere specks, though they may be numerous."

Both Internal Disease and Streak-disease are prevalent in Scotland. Most of the samples of seed potatoes affected with Streak-disease, purchased from dealers in Northumberland, were raised in Scotland, and affected tubers received from potato-growers in Durham county could be traced to Scotch seed. One correspondent sends the information that "sprain" is as bad in Scotland as Black Scab is in England but it is difficult to understand how much is included under the term "sprain" in statements of this kind.

A short time ago, Dr G. H. Pethybridge sent me a description of samples of potatoes affected with "rusty brown spots" which he had received from potato-growers in Ireland. A potato was sent for examination so that the disease could be diagnosed as a case of Streak-disease.

Storage.

According to Frank, "Buntwerden" does not spread during the storage period. In order to test this point a number of potatoes were cut in half, sprinkled with lime and kept through the winter under suitable conditions. Four lots of "The Sutton Flourball" variety were chosen—1, selected free from Internal Disease; 2, badly affected;

3, slightly affected and 4, very slightly affected. At the end of April, 1910, the potatoes in lot 2 were dried-up and shrivelled; those in lots 3 and 4 were not more badly diseased, and those in lot 1 remained free from disease with the exception of two or three halves which were slightly spotted. A similar result was obtained in the case of Streak-disease—the markings did not appear to spread in affected tubers and did not make their appearance in those selected free from disease. Potatoes from a sack affected with Phytopthora and bacteria were kept at the same time under similar conditions except that they were not sprinkled with lime—tubers selected free from Phytopthora and kept apart from the diseased ones did not develop disease, the disease did not spread in very slightly affected tubers, but the diseased ones were soon very badly affected with *Fusarium solani*. In the pit on the farm from which the potatoes came an additional loss was sustained owing to the ravages of Fusarium, the latter completing the mischief commenced by the joint incursion of Phytopthora and bacteria. Fusarium frequently appears in potatoes affected with Streak-disease and Internal Disease during the storage period, but I have kept several samples without the appearance of this fungus. The diseases in question are certainly not caused by *Fusarium solani*.

If the results obtained under experimental conditions do not agree with those experienced in the field they should not be too hastily regarded as contradictory, since the extent to which a particular disease contracted under one set of conditions (in the soil) spreads under a second set of conditions (in the pit or store) is not easy to determine owing to secondary ailments developed in the pit. These are apt to be confused with the former. Again, the extent to which disease does or does not spread during the storage period depends a great deal on the circumstance and condition of storing and keeping.

SUMMARY.

1. Internal Disease has been found to occur throughout numerous samples of potato of known variety such as for example: "The Sutton Flourball," "Sharpe's Express," etc.; similarly, Streak-disease has been found in samples of certain other varieties such as: "British Queen," "Edina," "Duke of York," etc.

2. Well-developed Internal Disease has been detected in very young tubers.

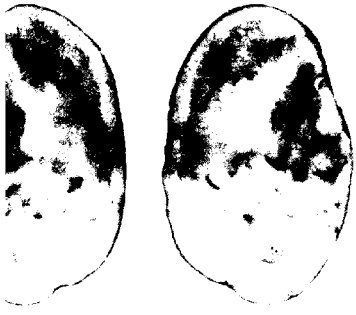
3. The markings in Internal Disease and Streak-disease may or may not form a connected system in the tissue of the potato, sometimes the discoloured areas are quite isolated. The diseased cells can be easily traced to within a few cells of the cork-layer (skin). In Streak-disease they can be occasionally traced right up to the cork or to a slight injury to the surface of the tuber.

4. No trace of a hyphal organism could be found either within the cells or intercellular spaces of diseased tissue. The cells are killed, often retaining their starch in an unaltered condition. If the disease is propagated from cell to cell it takes place without an easily visible deterioration of the cell-wall in the initial stages.

5. Internal Disease and Streak-disease did not spread in store under the experimental condition described.

6. In every experiment samples of potatoes affected with Internal Disease and Streak-disease whether planted in Devonshire or Northumberland produced a certain proportion of tubers affected with these diseases.

7. The diseases in question are frequently complicated by the presence of *Phytopthora infestans* in the field and *Fusarium solani* in the pit or store.



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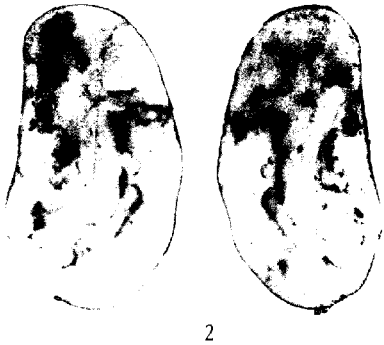

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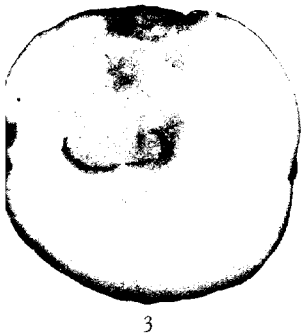

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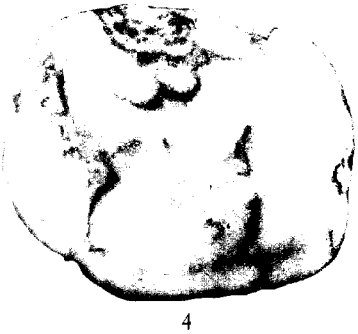

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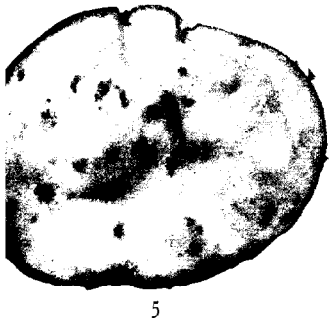

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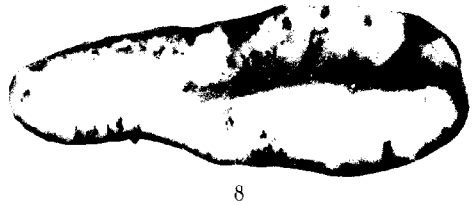

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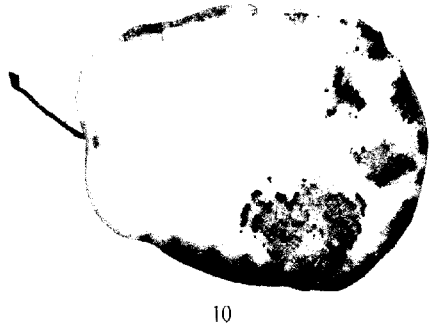

10

INDIAN INSECT LIFE, A MANUAL OF THE INSECTS OF THE PLAINS (TROPICAL INDIA).

By H. MAXWELL-LEFROY, M.A., F.E.S., F.Z.S.,

ASSISTED BY

F. M. HOWLETT, B.A., F.E.S.

W. Thacker & Co., London, 1909 (Price 30s.).

To those who for the first time visit a tropical country nothing is more striking than the extraordinary profusion of insect life. Some by their abundance, others by their bizarre form, others again by the brilliance of their colouring or through some suggestive vague resemblance to familiar northern forms, stimulate curiosity and excite a longing to learn more of their strange lives. This need the authors have attempted to satisfy in so far as it can be done within the limits of a single volume. Such a work must of course be sketchy, and probably no one realises this better than the authors themselves. Insects are already responsible for 14 volumes of the Fauna of British India and the tale is yet to run. The present work may be regarded in some ways as an introduction to that more comprehensive store of information, and Mr Lefroy has thoughtfully made cross reference easier by adopting the classification and nomenclature there in use. But the present work is more than a guide to the Fauna. Mr Lefroy has evidently had in his mind a volume which should appeal to the interested amateur. He has simplified classification, explained technical terms, and interspersed a wealth of illustrations in a way that cannot fail to interest and attract those whose only entomological equipment is that greatest gift of curiosity. The pictures form a feature of the book, for besides the 84 plates, of which the greater part are coloured, there are over 500 illustrations in the text. We think it unfortunate, however, that the heavily loaded type of paper has been employed. It adds unduly to the weight and bulk of the volume, and as most of the text illustrations are boldly drawn in black and white

there seems no reason why a lighter and thinner paper with a less highly glazed surface was not used. Perhaps this may be altered in another edition. Most of the plates appear to us to be excellent and we would particularly commend those illustrating Mr Howlett's account of the Diptera, which section, moreover, strikes us as being one of the best in the book. Some of the plates are not quite so happy. *Ergolis merione* among the butterflies has a washy look, while the figure on Plate 34 does scant justice to the viciously brilliant appearance of *Euchromia polymena*. But take it altogether it is a book which can be heartily recommended to all who take an intelligent interest in the insects of India, while at the same time, teeming as it does with the first-hand information of practical experts, it cannot but prove of the greatest service to all who are connected with agriculture in that large and important region of the earth.